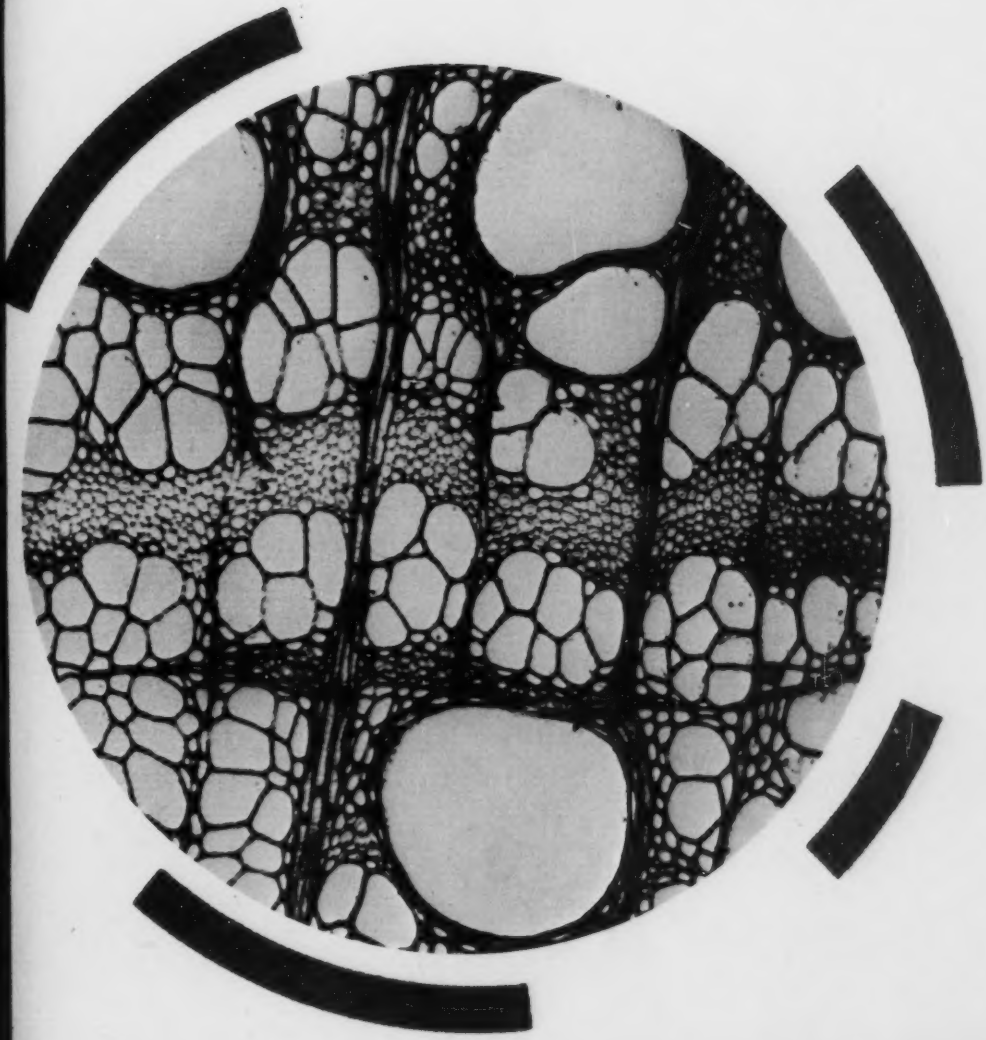
A Quarterly Journal of Research and Technical Progress

FOREST *Science*



december, 1956

PUBLISHED BY SOCIETY OF AMERICAN FORESTE

WASHINGTON, D.C

FOREST SCIENCE is published quarterly by the Society of American Foresters with the cooperation of forestry research and educational organizations.

The journal assumes no responsibility for the statements and opinions expressed by contributors. Although due care will be exercised in the handling of materials submitted for publication, the editors cannot be responsible for loss or damage of manuscripts and accompanying matter while in transit or in their possession.

Correspondence concerning manuscripts and other editorial matters should be addressed to Prof. Stephen H. Spurr, School of Natural Resources, University of Michigan, Ann Arbor, Michigan.

Other correspondence, including subscriptions and remittances, should be addressed to the Society of American Foresters, Mills Building, Washington 6, D. C.

Subscription price is $6.00 per year for complete volumes (one per calendar year), and $2.00 for single number. Add $0.25 for postage to Canadian addresses and $0.50 for postage to other foreign addresses.

The pages of FOREST SCIENCE are open to all qualified contributors. Preference will be given to papers devoted to the presentation and discussion of the results of original research. The voluntary payment of publication costs is encouraged. Authors are expected to bear the cost of that portion of their papers in excess of 16 printed pages in length and of tabular and illustrative material constituting more than 20 percent of the length of the paper. See suggestions on inside of back cover.

Board of Editor

Stephen H. Spu

Edito

School of Natural Resour

University of Michig

Henry Clepp

Society of American Foreste

V. L. Harp

Forest Service, U. S. Department of Agricultu

Business Manag

Miss L. Audrey Warn

Society of American Foreste

Copyright, 1956, by the Society of American Foreste

Entered as second-class matter at Washington, D. C., and Baltimore, Md.

Printed in the United States of Americ

FOREST *Science*

A Quarterly Journal of Research and Technical Progress

Suggested modifications of the standard Douglas-fir site curves for certain soils in southwestern Washington
Willard H. Carmean 242

Controlled pollination techniques for the Douglas fir
A. L. Orr-Ewing 251

Effects of temperature upon the activity and the behavior of the Douglas fir beetle
J. A. Rudinsky, J. P. Vite 258

Nitrogen loss from granitic soils on cheatgrass brome ranges
Harold F. Haupt 268

The anthocyanin and leuco-anthocyanins of sugar pine seedlings
Stanley Krugman 273

Taxonomy of the Cenangium dieback fungus
Hugo A. Ferchau, T. W. Johnson, Jr. 281

Sampling methods and community attributes in forest ecology
Alton A. Lindsey 287

Tissue differentiation and the application of sodium arsenite to girdled trees relative to bark peeling
M. Reines 297

Measurement of acorn crops
Leslie W. Gysel 305

A spherical densiometer for estimating forest overstory density
Paul E. Lemmon 314

Germination of slash pine pollen in vitro
R. M. Echols, Francois Mergen 321

Photoperiod and flowering of pines
N. T. Mirvov 328

Index to Volume 2 333

REVIEWS: Forest and range policy 250, Forestry and wildlife atlas for Czeckoslovakia 257, Argentine woods and forests 285.

The cover photomicrograph is of American elm in transverse section by Everett Ellis. That in the June Issue was of white oak rather than elm as stated.

Volume 2
December 1956
Number 4

Suggested Modifications of the Standard Douglas-fir Site Curves for Certain Soils in Southwestern Washington

BY
WILLARD H. CARMEAN

CONVENTIONAL site index curves for even-aged forest stands are usually based on the heights and ages of dominant and codominant trees measured within a particular area or region. Data for these curves are obtained from sample plots established in well-stocked, even-aged stands that occur within the range of ages and sites found for the species being studied.

The total heights and total ages of dominant or of dominant and codominant trees found in the sample plots are used to calculate an average site index curve. Site curves for a range of sites above and below the average curve are then calculated as percentages of the average curve. The mensurational procedure for the construction of site index curves has been described by Bruce (1926) and Bruce and Schumacher (1950).

In conventional site curves it is assumed that the sample plot data represent a normal distribution of site quality within each age class. It is possible, however, that the average site curve used in calculating curves for good and poor sites will be biased if certain age classes contain a preponderance of either good or poor site plots. Mensurational procedures have been developed (Bruce and Schumacher, 1950) for altering the shape of the curves for good and poor sites calculated from the initial average site curve. These procedures, however, will not correct for bias that might exist in the guiding site curve itself.

Because the conventional method is based on a single average site curve, the shape of the site index curves (i.e., the rate of height growth) will be assumed to be the same for all site classes, localities, and soil types that were included in the basic site curve study. The average site curve, however, may not be applicable to certain site classes, localities, or soil types that were not adequately represented by sample plot data. Serious errors in estimating site index may occur if the conventional site curves characteristic of the average conditions found in the site study differ in shape from site curves that would be applicable to certain unusual stand and soil conditions. Even within specific soil types or specific localities it is possible that the shape of the site curves for good-site-quality stands might differ from that of the curves for stands of poor site quality.

Foresters who have made periodic measurements of permanent sample plots have occasionally observed that site index apparently changes as forest stands grow older.

The author is Soil Scientist, Central States Forest Experiment Station, Forest Service, U. S. Department of Agriculture. This study was made while the author was on the staff of the Pacific Northwest Forest and Range Experiment Station, Portland, Ore. Appreciation is extended to Professor F. X. Schumacher, Duke University, Durham, N. C., who suggested the statistical procedure. The Washington State Division of Forestry provided assistance in the collection of field data.

TABLE 1. Coefficients of the age variable on the major soil groups of southwestern Washington.

Soil group	Number of plots	Coefficient of the age variable		Standard error
Gravels[1]	43	— 5.122		±3.808
Sands[1]	19	— 9.152		±4.336
Basalts and basalt conglomerates	44	—13.737	—13.832	±1.330
Shales and sandstones	24	—13.960		±0.908
Old valley terraces and glacial, lacustrine deposits	25	—20.241		±2.891

[1]The coefficient for sands is not significantly different from those for the other soil groups; therefore, site curves have not been prepared for them. The curve for sands in Figure 1 is presented only for comparison. The coefficient for gravels is significantly different from the combined coefficient for basalts, shales, and sandstones, but not from zero. The null hypothesis that the coefficient of the age variable is zero is, however, untenable for obvious reasons.

Such observations have been reported by Lange (1951) and by Watt (1953). It usually has been difficult to decide whether the apparent change in site index is due to an actual change in site quality or to errors in the site curves from which site index is estimated. Unless a severe disturbance occurs, such as heavy grazing, intensive burning, erosion, drainage, or flooding, it appears unlikely that any great change occurs in the inherent productive capacity (i.e. site quality) of forest areas where soil conditions have reached a relative equilibrium under centuries of natural soil development. Accordingly, any consistent sustained change in site index measured on permanent plots probably indicates errors in the site curves (Spurr, 1952, 1956).

Soil-site studies of pine in the Southeast have produced methods that can be used in the construction of site curves. These curves are statistically independent of the environmental factors found to be related to the height growth of dominant and codominant trees and are more nearly a true expression of the relationship that exists between tree height and tree age. Coile and Schumacher (1953) and Gaiser (1950) have used these methods for correcting conventional site curves for loblolly pine (*Pinus taeda* L.) in the Southeast.

The Study

A study of the soil-site relations of Douglas-fir (*Pseudotsuga menziesii* (Mirb.) Franco) in southwestern Washington provided data for classifying site quality on forest land in this area independent of the type or condition of the existing vegetal cover (Carmean, 1954). This study also provided data that could be used to construct local site curves for Douglas-fir growing on important soil groups found in the same area. These soil-site curves can be used to appraise the local applicability of the site curves commonly used for Douglas-fir in the Pacific Northwest (McArdle, 1949).[1]

The soil-site study consisted of an analysis of Douglas-fir site quality on five major soil groups in southwestern Washington. Data were taken on 155 circular one-fifth-acre plots established in well-stocked, even-aged, undisturbed young Douglas-fir stands. Though tree ages ranged from 28 to 108 years, most plots were in stands 40 to 70 years of age. The total age and total height of six dominant trees on each plot were recorded, along with a description of the soil and topographic conditions. Samples of all soil horizons were taken for laboratory analysis. The number of plots established on each of the five soil groups is given in Table 1.

Multiple regression equations that express total dominant tree height in terms of

[1]The curves developed by McArdle are referred to subsequently in this paper as the standard site curves.

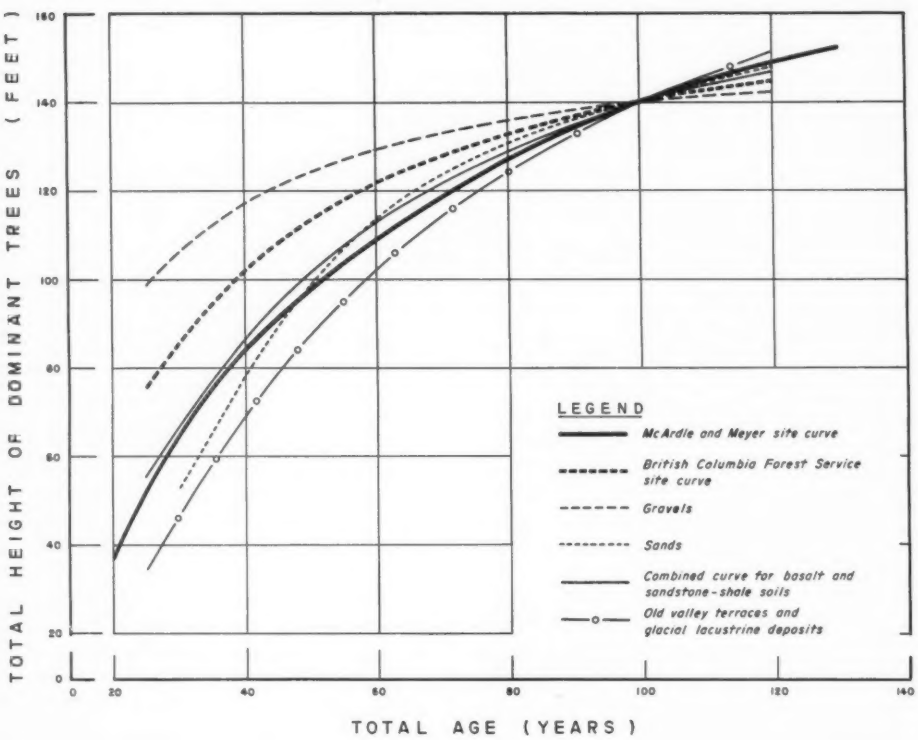


FIGURE 1. *Douglas-fir site index 140 curves for young, well-stocked even-aged stands growing on the major soil groups of southwestern Washington. The standard curve and the British Columbia Forest Service site index 140 curve are also shown for comparison. The soil-site curves express the change of tree height with age for various soil groups when site index is held constant at 140. They do not represent average site quality for these soil groups.*

age and associated factors of soil, precipitation, and topography were calculated for each soil group. Results of this analysis showed that total tree height was related to total tree age for all soil groups and that tree height was also related to precipitation, elevation, and certain soil conditions.

Results varied with soil groups as follows:

1. With gravelly glacial and alluvial soils, increased site quality was related to decreased elevation and gravel content and to increased annual precipitation.

2. For sandy glacial and alluvial soils, increased site quality was correlated with decreased elevation and gravel content and with increased effective soil depth.

3. For residual soils derived from basalts and basalt conglomerates, site quality increased with decreases in elevation and increases in the product of moisture equivalent and gravel content.

4. For residual soils derived from shales and sandstones, increased site quality was related to increase in effective soil depth.

5. For imperfectly drained soils derived from old valley terraces and glacial lacustrine deposits, increased site quality was related to decreased elevation and gravel content, increased annual precipitation, and improved internal drainage.

Regression coefficients of the age variable, independent of the effect of the factors of soil, precipitation, and elevation, are given in Table 1 for each of the soil groups listed above. In comparison with older methods, the calculation of coefficients in this way is believed to provide a more exact expression of the true relationship between tree age and tree height, because the confounding effects of tree environment have been statistically minimized.

The coefficients in Table 1 were used in the following equation to calculate the site curves in Figure 1.

$$\text{Log } H = \log \text{S.I.} + b_1\left(\frac{1}{x_1} - 0.01\right), \text{ where}$$

log H = logarithm of total dominant tree height
log S.I. = logarithm of site index
b_1 = coefficient of the age variable
x_1 = total age in years

Curves for site index 140 (tree height at 100 years) were calculated for all soil groups, since this is the average site index of the standard Douglas-fir site curves. They also provide a comparison of the shape of all site curves at the same site index value.

Results and Discussion

The standard site index 140 curve compares favorably with the soil-site curve for the combined basalt and shale-sandstone soil groups (Fig. 1). This suggests that reliable site index values are obtained when the standard site curves are applied to stands growing on the residual upland soils of southwestern Washington. Probably a large number of the yield table plots were located on soils derived from basalts, andesites, shales, and sandstones, for these soil groups occupy very extensive areas in the entire Douglas-fir region as well as in southwestern Washington (U. S. Department of Agriculture, 1938).

A recent study by Lemmon (1955) on Douglas-fir site quality in the Willamette Basin of Oregon indicates that stands established on the residual soils of this area have height growth similar to that of stands growing on the residual soils of southwestern Washington. Lemmon used analytical procedures similar to those of the present study and obtained coefficients for stands growing on soils derived from basalt, shale, and sandstone that differed very little from the coefficients obtained in the present study (Table 1). These results suggest that Douglas-fir height growth on the residual soils in southwestern Washington and in the Willamette Basin follows the pattern presented in the standard Douglas-fir site curves.

Site curves for soils of coarse texture (gravels and sands) and for soils of imperfect internal drainage (old valley terraces and glacial lacustrine deposits) have a different shape than the standard site curves. Early height growth of Douglas-fir is relatively rapid on gravels and sands but tends to level off after 50 years of age (Fig. 1). This accords with the general observation that the height growth of stands growing on certain gravelly glacial soils appears to "stagnate" at about 60 to 70 years. Many old-growth trees on these soils are not much taller than trees found in much younger "second-growth" stands. Figure 1 also indicates that trees growing on imperfectly drained soils have a relatively slow rate of early height growth, but after 100 years of age they do not show the pronounced slowing of height growth which is characteristic of trees on coarse textured, better drained soils.

A comparison of the shapes of various Douglas-fir site curves has been made by Spilsbury and Smith (1947). This study compared site curves from Spilsbury and Smith's plant indicator study, the curves used by the British Columbia Forest Service (1936), and the standard Douglas-fir site curves. The B.C.F.S. curve is steep for the first 50 years; subsequently a slowing of height growth occurs (Fig. 1). Although the B.C.F.S. curve is similar in shape to those for gravels and sands, it shows less difference between the growth rates at early and later ages. The Spilsbury

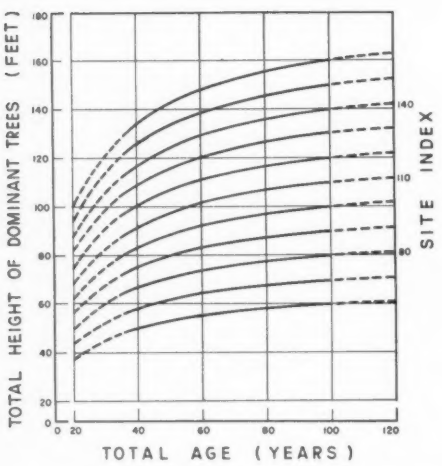


FIGURE 2. *Modified site index curves for young, well-stocked, even-aged Douglas-fir stands growing on gravelly soils of southwestern Washington. Applicable to soil series: Everett, Grove, Fitch, Spanaway, Deckerville, Alderwood, Hoodsport, Sinclair, Shelton, Nasel, Giles (gravelly phase).*

and Smith vegetative site type curves generally agree with those of the British Columbia Forest Service. It is possible that the difference in shape between the B.C.F.S. curves and the McArdle curves can be attributed to differences between the soils of British Columbia and those of Oregon and Washington.

The site curves (for site index 140) in Figure 1 indicate that large errors may result if the standard Douglas-fir site curves are used for estimating the site index of trees growing on gravels, sands, and imperfectly drained soils. When dominant trees are much less than 100 years of age, site quality will be overestimated for Douglas-fir growing on gravels and sands, because trees that are site index 140 (standard curves) at young ages will not be able to maintain the height growth predicted by the standard site index curves. Hence, these young stands will appear to decrease in site index as their age approaches 100 years.

For imperfectly drained soils (old valley terraces and glacial lacustrine deposits) the standard site index curves will give underestimates of site index. On these soils rapid height growth is maintained for a larger period of time than is predicted by the standard site curves. Thus the site index of stands growing on imperfectly drained soils will apparently increase as the stands approach 100 years of age.

An example of the type and magnitude of errors that may occur in site index estimation is as follows. If several dominant and codominant trees growing in a normally stocked stand on gravelly soil have an average total height of 84 feet at 40 years, a forester using the standard site curves would estimate the site index to be 140 feet at 100 years. Using the yield tables (McArdle, Table 3) he would estimate the present volume at 22,000 board feet per acre (International 1/8″ Rule, trees 7″ and larger). There would be no error in estimating present volume, assuming that the yield tables apply to all Douglas-fir stands regardless of the soil conditions.[2]

A serious error can be expected, however, if the forester uses the standard site curves to predict future site index and future volume of this stand. If trees on gravels grow in height as predicted in Figures 1 and 2, then trees 84 feet tall at 40 years might actually be only 102 feet tall when they reach 100 years of age. A second forester examining this stand when it is 100 years old would estimate the site index to be 102 feet (standard curve) and by interpolation in the stand-

[2]Standard yield tables and site curves are currently used for all Douglas-fir stands regardless of the soil conditions involved. The present study indicates that modified site curves are needed for soils with excessive or imperfect internal drainage. It may be logically expected that future study will reveal a further need for modified yield tables for these soils. Until such studies are completed, however, there is no simple alternative to using the standard yield tables, in which all soil and stand conditions are averaged.

ard yield tables would record the volume as 48,980 board feet per acre.

In this example the first forester overestimated future height and site index by 38 feet (140 — 102) when he used the standard curves at age 40. Furthermore, he predicted that the volume at 100 years would be 94,700 board feet per acre, with site index 140. He thus overestimated future volume at 100 years by 45,720 board feet per acre (94,700 — 48,980), or 93 percent.

In the example given, the forester correctly used the standard site curves at age 40 to identify present site index and present volume. If he wishes to identify future site index and future volume on gravelly soils, however, he should use the soil-site curves in Figure 2 to estimate tree height at specific future ages of this stand.

It is apparent that an owner of large areas of timber occurring on the gravelly or the imperfectly drained soils of southwestern Washington may make serious errors in estimating future Douglas-fir yields if his site index estimates are faulty, as in the example above. These errors might seriously affect sustained yield management plans and, where future growth and yield are greatly overestimated, could result in overcutting.

Modified Douglas-fir Site Curves for Certain Soil Groups of Southwestern Washington

Local site curves for gravels and also for imperfectly drained soils are given in Figures 2 and 3. These modified site curves can be used with the standard site curves and yield tables to estimate future height and volume of Douglas-fir stands on these soils. The modified curves may enable foresters to obtain more precise estimates of site and volume than seem possible with present standard site curves alone. Additional studies may be required to obtain more reliable information on growth and yield on these soils than is now available.

The site curves in Figures 2 and 3 are based on the coefficients listed in Table 1. They were calculated by the same methods as the curves in Figure 1.

No curves are given for stands growing on sandy glacial and alluvial soils, because the standard error indicates that much variation is associated with the coefficient for this soil group (Table 1). However, for stands growing on sandy soils, the site curves for gravels may be used if the site quality appears poor, and the standard site curves if the site quality appears good. No site curves were calculated for the combined basalt and shale-sandstone soil groups, since no significant differences exist between the standard site curves and the curves of these soil groups (Table 1 and Fig. 1).

The curves in Figures 1, 2, and 3 are based on data taken from dominant trees only. It is believed, however, that the modified curves can be used with tree measurements based on either dominant trees or on dominant and codominant trees as used in the yield study. Staebler (1948) found that the combined heights of dominant and codominant Douglas-fir trees had a linear relationship to the height of dominant trees. Although dominant trees are somewhat taller than codominant trees, Staebler's study indicates that the shapes of the height curves for the two classes of trees are similar. It may therefore be assumed that changing proportions of dominant trees to codominant trees with increased stand age will have relatively little effect on the shape of the curve for combined dominant and codominant tree height growth.

Staebler also found that dominant tree height can be reduced to the combined height of dominant and codominant trees by the expression:

$$\text{Diff.} = 2.66 + 0.040\ (H_D), \text{ where}$$

Diff. = Difference between the average height of dominants and codiminants.

H_D = Average height of dominant trees.

Until further information becomes available, the following is suggested as a procedure for measuring present and future

site index and volume of young Douglas-fir stands growing on gravels or on imperfectly drained soils of southwestern Washington.

a. Measure the present total height and total age of five or more trees. Present site index and volume can be estimated by relating average total tree height and age to the standard site curves and yield tables. If dominant and codominant trees are measured, a direct comparison to the standard site curves can be made using the customary height over age methods. It is suggested, however, that greater accuracy will result if measurements are taken only on dominant trees.[3] For comparability, the dominant tree height must then be somewhat reduced as previously indicated.

b. An estimate of future tree height and site index can be made by relating present average tree height and age values to the appropriate modified soil-site curve (Figs. 2 and 3). Future tree height at specific ages can be estimated by following the selected site index line to its intersection with the ordinate for the given age, and reading from the height ordinate (Y axis) of the site curve.

c. Future tree height at the given future age is then related to the standard site curves to obtain a future site index value that is directly comparable to the Douglas-fir yield tables. This future site index value is used to estimate stand volume at the given future age.

For example, if several dominant and codominant trees growing on gravelly soil have an average total height of 84 feet at 40 years, then present site index, using the standard site curves, is 140. If 84 feet is the average height of dominant trees only, however, this value should be adjusted to an average of 78 feet for dominants and codominants by use of Staebler's correction equation. With the adjusted height of 78 feet at 40 years, the present site index is 130, according to the standard curves. An estimate of present volume can be obtained from the standard yield tables by relating site index 130 to stands 40 years of age.

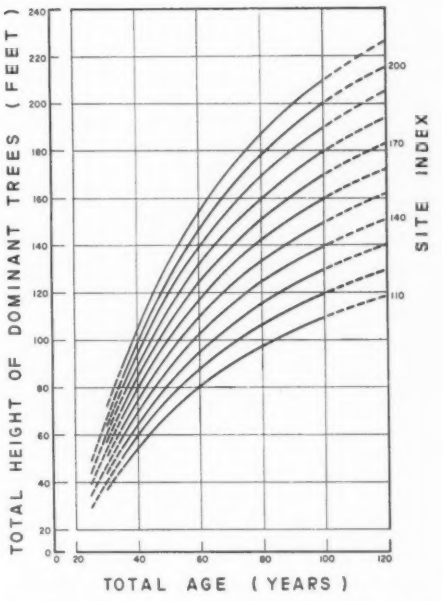


FIGURE 3. *Modified site index curves for young, well-stocked, even-aged Douglas-fir stands growing on southwestern Washington soils derived from old valley terraces and glacial lacustrine deposits (soils with imperfect internal drainage). Applicable to soil series: Kitsap, Cloquallum, Saxon, Salkum, Scammon, Kopiah, Winlock, Lacamas, Olequa, Grand Ronde, Wapato, Chehalis, Stevenson.*

An estimation of future tree height, site index, and volume requires the use of the modified curves in Figure 2. As before, tree heights should be in terms of dominant and codominant trees so that estimates of future tree height can be properly related to the standard site curves and yield tables. An average height of 84 feet for dominants

[3]Ker (1952) has shown that the combined data of dominant and codominant trees are much more variable than data taken from dominant trees alone. Hence if dominant trees only are measured, fewer are needed to obtain the same accuracy of estimate. Also, dominant trees are easier to classify and measure, and no consideration need be given to the ratio of dominant to codominant site trees.

and codominants at 40 years of age coincides with the soil-site curve for site index 102, or a height of 102 feet when this stand reaches 100 years of age. The height ordinate of the same soil-site curve shows that these trees will average 99 feet in height at age 80. This value is then related to the standard site index curves to obtain a standard site index of 109 feet. By interpolation in the standard yield tables, the estimated volume of a site index 109 stand at 80 years is 44,760 board feet per acre (International 1/8″ rule, trees 7″ and larger).

Summary

Douglas-fir site curves for trees growing on five major soil groups in southwestern Washington have been calculated from data gathered in a recent Douglas-fir soil-site study. It was found that site curves for stands growing on soils derived from basalts, sandstones, and shales are similar to the standard Douglas-fir site curves used in the Pacific Northwest. However, trees growing on gravels and sands and on soils with imperfect internal drainage (soils derived from old valley terraces and glacial lacustrine deposits) have markedly different site curves. Results indicate that when stands are less than 100 years of age the standard curves overestimate site quality on sands and gravels and underestimate site quality on imperfectly drained soils.

Modified site curves are presented for Douglas-fir in southwestern Washington established on gravels and on soils with imperfect internal drainage. Results of this study indicate that it would be desirable to have additional yield table and site curve studies for stands growing on these soils. Future studies should include more observations and a wider range of ages than were covered in the present study. In addition, the shape of curves for different site classes within specific soil groups might be considered.

Literature Cited

BRITISH COLUMBIA FOREST SERVICE. 1936. Volume, yield, and stand tables for some of the principal timber species of British Columbia. Victoria, B. C.

BRUCE, D. 1926. A method of preparing timber yield tables. J. Agric. Res. 32: 543-557.

———— and F. X. SCHUMACHER. 1950. Forest mensuration. 3rd Edition. McGraw-Hill Book Co., N. Y. 403 pp.

CARMEAN, W. H. 1954. Site quality for Douglas-fir in southwestern Washington and its relationship to precipitation, elevation, and physical soil properties. Proc. Soil Sci. Soc. Amer. 18: 330-334.

COILE, T. S. and F. X. SCHUMACHER. 1953. Site index of young stands of loblolly and shortleaf pines in the Piedmont Plateau Region. J. For. 51: 432-435.

GAISER, R. N. 1950. Relation between soil characteristics and site index of loblolly pine in the Coastal Plain Region of Virginia and the Carolinas. J. For. 48: 272-275.

KER, J. W. 1952. An evaluation of several methods of estimating site index of immature stands. For. Chron. 28: 63-74.

LANGE, K. D. 1951. Effects of clearcutting understory hardwoods on the growth of a shortleaf-Virginia pine stand. J. For. 49: 176-178.

LEMMON, P. E. 1955. Factors affecting the productivity of some lands in the Willamette Basin of Oregon for Douglas-fir timber. J. For. 53: 323-330.

MCARDLE, R. E. 1949. The yield of Douglas-fir in the Pacific Northwest. U. S. Dept. Agric. Tech. Bull. 201. 74 pp. Revised.

SPILSBURY, R. E., and D. S. SMITH. 1947. Forest site types of the Pacific Northwest; a preliminary report. Brit. Columbia For. Serv. Tech. Pub. T 30. 46 pp.

SPURR, STEPHEN H. 1952. Forest inventory. The Ronald Press Co., New York. 476 pp.

————. 1956. Soils in relation to site-index curves. 1955 Proc. Soc. Amer. For. Meeting. pp. 80-85.

STAEBLER, G. R. 1948. Use of dominant tree heights in determining site index for Douglas-fir. U. S. For. Serv., Pacif.

Nthwest. For. Range Exp. Sta., For. Res. Note 44. 3 pp.
U. S. DEPARTMENT OF AGRICULTURE. 1938. Yearbook, Soils and Men: 1045. U. S. Govt. Printing Office, Washington, D. C.
WATT, R. F. 1953. Site index changes in western white pine forests. U. S. For. Serv., Nth. Rocky Mt. For. Range Exp. Sta., Res. Note 132. 2 pp.

Forest and Range Policy

Its development in the United States. By Samuel Trask Dana. McGraw-Hill Book Co., Inc. New York. 455 pp., 1956. Price $6.50.

Review by Stephen H. Spurr
University of Michigan, Ann Arbor, Michigan

Probably more books have been written about forest policy and the history of our public domain than about any other phase of American forestry. Few if any, however, have combined broad coverage of the field with objective presentation and careful array of facts. Dean Dana's book, the second on the subject (after Col. Greeley's) published in the American Forestry Series, is perhaps the first to near this goal.

Forest policy may be approached from an historical viewpoint or from a classification of major issues. Dean Dana has chosen the former, and his book with equal accuracy could have been titled a history of American forestry. In the eleven chapters that precede the summary, he narrates the public and private policies and practices from early colonial days to the present. The reader is soon struck with the brevity and preciseness with which this history is unravelled. Seventeen pages carry us from the Pilgrims to the Revolution, five pages summarize the Ballinger affair, and two the C.C.C. In fact, the writing becomes a little choppy at times because of the brevity of treatment of one important topic after another. The reader soon finds, though, that each page is so full of vital facts and carefully weighed sentences that he can profit best from reading slowly and even working his way through only a few pages at a time.

Only as the book is digested does one become aware that the survey of American forest policy is not only encyclopediac, but also exceptionally well balanced and remarkably objective. Each happening and each issue is in its place. All sides of the problem are aired. Even the devil is given his due in a few properly chosen sentences whenever he enters the scene—which has apparently occurred frequently in our forest history.

It is inevitable that such a concise coverage of a big topic should have its omissions—and a noteworthy one is the complete absence of the author himself in the entire text. Sam Dana has been far more than a teacher of forest policy and the respected dean of a forestry school (University of Michigan, 1927 to 1951). Perhaps more than any other American forester of our generation, he has become known for his ability to chair critical forestry and conservation meetings and for the wisdom and objectivity of his own advice. In short, Dean Dana himself has been a key force in forming American forest policy and should be recognized as such, even in his own book.

The appendices cover more than 80 pages. The first summarizes federal policy in wildlife, soil, water, and minerals—fields related to forest and range policy. The second is a detailed chronological summary of important events in the development of Colonial and Federal policies relating to natural resources. It is a reference source of the highest value.

Forest and range policy is written as a text and for reference purposes. In treatment, objectivity, and authority, it fully meets the requirements for this type of book and should find extensive adoption in our colleges and professional schools. It should also prove a valuable reference work for the professional forester and for the conservationist generally.

Controlled Pollination Techniques for the Douglas Fir

BY
A. L. ORR-EWING

THE CONTROLLED BREEDING of coniferous species presents certain problems which are somewhat different to those encountered in the breeding of agricultural crops. The seed production habits of most conifers are sporadic and faulty breeding techniques can seriously delay a programme of seed improvement. The following information on controlled pollination techniques for the Douglas fir (*Pseudotsuga menziesii* (Mirb.) Franco. was obtained during a recent cytological investigation of this species.[1]

Isolation of the Female Cone

The most important prerequisite in the controlled pollination of conifers is that the female cones be effectively isolated from any exterior pollen contamination so it is essential that their development in early spring is carefully checked. The Douglas fir is monoecious and the microsporangiate and megasporangiate strobili, subsequently referred to as the male and female cones, are often found together on the same branchlet. Figure 1 shows typical male and female cone buds of the Douglas fir in late February. The female buds can be readily distinguished as they are both larger and more pointed than the male buds and occur laterally to the terminal vegetative bud. Figure 2 shows a more advanced stage of development, the male buds are now considerably swollen and the cones will shortly emerge. The female cone buds should be isolated at this stage and before that shown in figure 3, where the male cones are almost mature and the female cones are emerging. Isolation as late as this stage would involve some risk of contamination because pollen from more precocious trees could now settle on the female cone. Table 1 shows the dates on which the female cone buds of the eight trees in the investigation were isolated. Trees 1 and 2 were located at Vancouver and the remainder at Victoria and Lake Cowichan on Vancouver Island. There was considerable variation in the development of the male and female cones in trees from even the same locality. This development also varied on individual trees as both the male and female cones matured more rapidly at the top of a tree and on southerly aspects.

The female cone buds were isolated with bags constructed from viscose sausage casing as recommended by Duffield (1950)

[1]Orr-Ewing, A. L., 1956. An investigation into the effects of self-pollination on *Pseudotsuga menziesii* (Mirb.) Franco. Diss., University of British Columbia, Vancouver.

British Columbia Forest Service Publ. T. 44. The author is a member of the Research Division of the British Columbia Forest Service, Victoria, B. C., Canada. He expresses his appreciation of the photographic assistance by R. Banyard of the Forest Biology Division, Canada Department of Agriculture, Victoria, B. C.

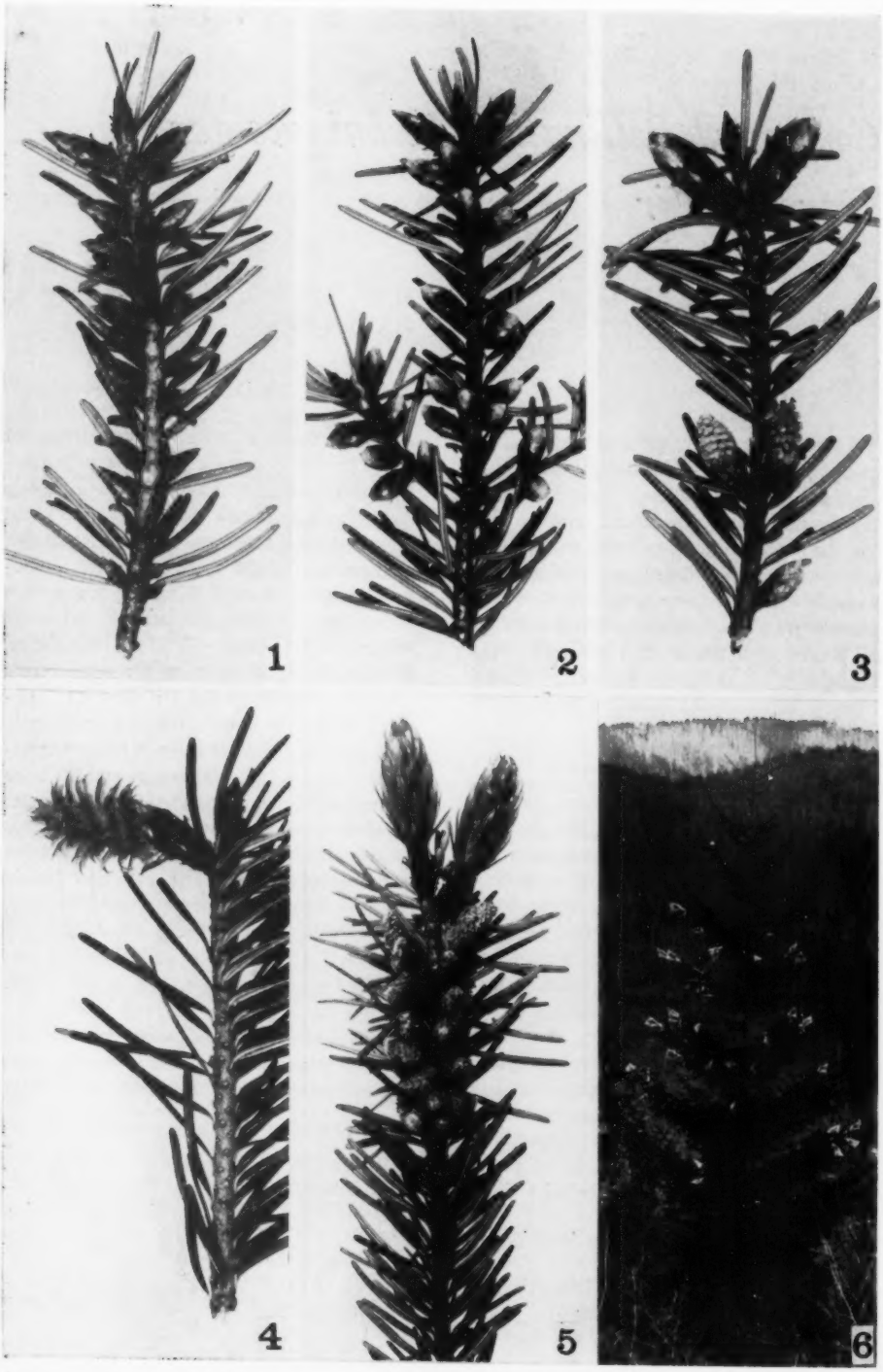

1
2
3
4
5
6

TABLE 1. Limits of isolation and pollination of cones.

Tree	No. of bags	No. of cones isolated	Dates of isolation	Dates of pollination	Dates of bag removal	No. of cones collected
1[1]	47	100	Mar 11-15	Mar 30-Apr 7	Apr 27-28	74
2	31	105	Mar 15-23	Apr 14	Apr 27	76
A	73	310	Mar 22-Apr 13	Apr 16-May 3	May 1-9	224
C	57	202	Mar 25-Apr 6	Apr 13-16	Apr 24-May 3	104
D	71	271	Mar 26-Apr 10	Apr 15-26	May 4-8	126
F	47	280	Mar 29-Apr 20	Apr 29-May 5	May 14-15	212
H	58	278	Mar 31-Apr 20	Apr 21-29	May 14	179
J	48	268	Apr 1-2	Apr 21-29	May 13-14	220

[1]Trees 1 and 2 were pollinated in 1952, the remaining trees in 1954.

and one of these bags is shown in figure 8. The female cones have been left ample space in which to develop and the bag has been securely tied around absorbent cotton to the branchlet. Care should be taken to remove all the male cone buds from the portion of the branchlet which will be enclosed with the female cone or they will mature and release pollen. The date of isolation and the number of female cone buds enclosed in each of the numbered and tagged bags are then recorded in the special form shown in figure 7 which was developed from the one described by Cumming and Righter (1948). These viscose casing bags were found quite satisfactory as they are cheap, light, and transparent and can withstand the variable spring weather. They also effectively screen the female cones from any pollen contamination provided that they are correctly constructed and used at the right stages of development. Their one disadvantage is that heat is absorbed more readily than by other types of bags with the result that some mortality of the young cones must be anticipated. Nienstaedt and Kriebel (1955) have found that the internal temperature within this type of bag was high compared with that within other types of bags. Such high temperatures can kill the young cone, either prior to or shortly after emergence. This mortality, however, can be substantially reduced by restricting the isolation of cones to the northerly sides of the tree as far as possible.

Pollination

Pollen must be available at the time when the isolated female cones are most receptive to pollination so that it should be collected early in the spring as the cones develop rapidly within the bags. A satisfactory method for extracting Douglas fir pollen has already been described (Orr-Ewing, 1954). Branchlets with male cone buds are removed and brought indoors in early March. They are placed in water at room temperature and pollen can be readily collected when the cones mature. The pollen is then stored at 4°C. until required. This technique was found very satisfactory but pollen from different trees should be extracted in separate rooms.

In the meantime, the development of the isolated female cones must be carefully

FIGURE 1. *The male and female cone buds of the Douglas fir in early February. Victoria, 1954.* FIGURE 2. *The male buds are now considerably swollen and the female cone buds should be isolated.* FIGURE 3. *The male cones are almost mature and the female cones are emerging.* FIGURE 4. *Pollination should take place at this stage.* FIGURE 5. *The cone scales have closed so that further pollination is not possible.* FIGURE 6. *A tree with the pollination bags in position.*

Tree No._____Location_____

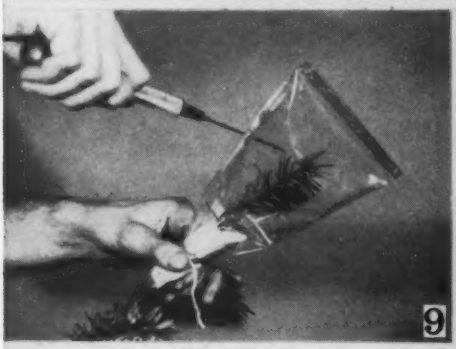
Flowers bagged____Bags changed_____

Cones bagged_____Cones collected____

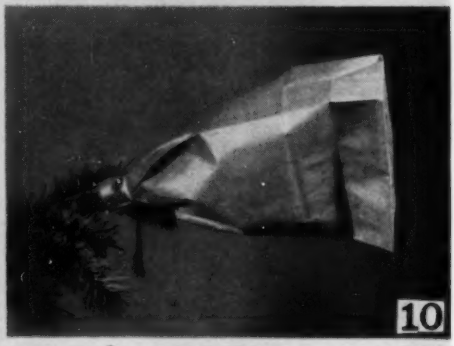
Tag No.	No. of Flowers	Pollen Parent	Date Bagged	Date Pollinated	Samples Removed	Cones Collected

7

8

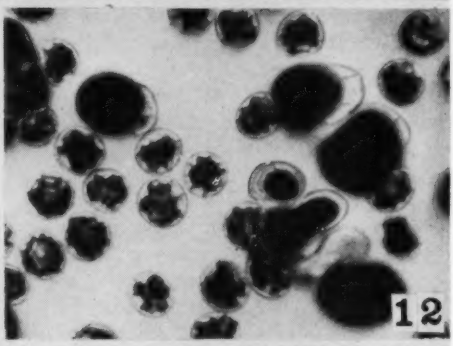


9

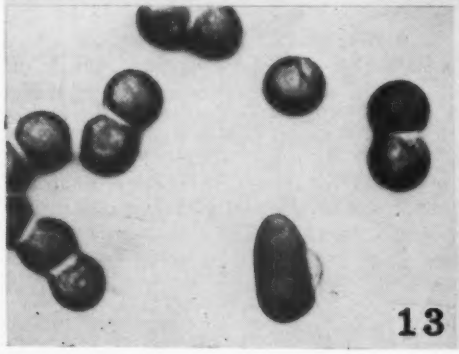


10

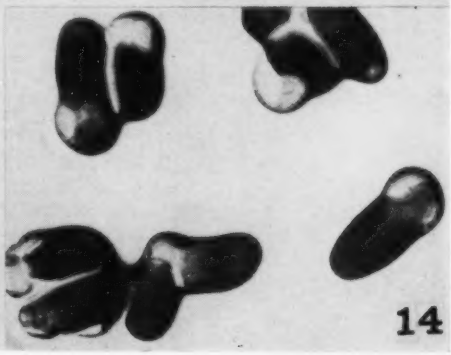
11



12



13



14

checked. The stage shown in figure 4 was considered to be the most receptive to pollination as the cone has completely emerged and the ovuliferous scales are fully extended.

Pollinations are made with a hypodermic syringe which is fitted with a rubber bulb and pollen agitator as recommended by Duffield (1950). The tip of the needle is pushed through the bag as shown in figure 9 and a cloud of pollen blown directly onto the female cones. The small hole in the bag is then sealed with tape. The pollen parent and date of pollination are next recorded on the special form and a coloured chicken ring slipped onto the branchlet at the base of the bag to show that pollination has been completed. The colour of this ring indicates the particular pollen parent. Pollination on any one tree generally took several days to complete as the cones do not all reach the receptive stage simultaneously. The viscose casing bags are removed as soon as there is no danger of foreign pollination as they soon become too small for the rapidly developing cones. It was considered safe to remove these bags when the cone scales had sealed as shown in figure 5 and the cones had begun to invert. The cones, however, must be protected against insect and squirrel attack and are, therefore, covered immediately with kraft paper bags which have no adverse effect on development. One of these bags is shown in figure 10. The seams should be stapled and sealed with waterproof glue and the bags can then be used for several seasons.

Isolation and pollination of the cones of a vigorous young tree, such as that shown in figure 6, is by no means any easy operation. A safety belt and light wire hook are indispensable for this type of work.

Pollen Germination

In any tree breeding programme, pollen should always be tested prior to use in order to ensure that it is viable or the results of controlled pollinations will be confused. No previous germination studies had been carried out with pollen of the Douglas fir and the following outlines the procedure found the most satisfactory. A solution of two percent bacto-agar was heated and then poured into a petri dish and allowed to cool. Pollen was then scattered lightly in five different places on the surface of the agar and a few drops of water were placed on the inside of the lid in order to maintain a high humidity. The petri dish and contents were then incubated for twenty-four hours at 26°C. At the end of this period, the five areas of pollen were examined under a low power microscope. A field of pollen was selected at random in each area and the amount of germination checked in fifty pollen grains. Figure 11 shows typical pollen grains of the Douglas fir. These are wingless, about eight microns in width and have a characteristic depression on the one side which is probably an adaptation for wind dissemination. Pollen was considered to have germinated when it had swollen and ruptured the outer exine layer with resultant tube growth. Vigorously germinating pollen is shown in figure 14.

Pollen Storage

Pollen storage may often be necessary in tree breeding as it is not always possible to make controlled crosses the same year that pollen is collected. No previous studies had been made on the storage requirements of pollen of the Douglas fir and some exploratory work was carried out. The different pollen lots throughout this investigation were stored at 4°C. in lightly

FIGURE 7. *A tree breeding record form.* FIGURE 8. *A female cone isolated with a viscose casing bag.* FIGURE 9. *Controlled pollination.* FIGURE 10. *The kraft bag used as protection against insects and squirrels.* FIGURE 11. *Pollen grains of the Douglas fir.* × 75. FIGURE 12. *Low germination of pollen stored for two years.* × 75. FIGURE 13. *Low germination of pollen stored one year.* × 75. FIGURE 14. *Vigorous germination of pollen stored one year* × 75.

TABLE 2. Pollination with two-year-old pollen, 1954.

Tree	Pollen parent	Pollen[1] germin.	No. of cones collected	No. of cleaned seeds	Germin.	Cutting test of ungerminated seed
A	3	120/250	8	1	0	Seeds empty
	4	128/250	22	6	0	Seeds empty
D	2	54/250	1	0	0	.....
	5	191/250	5	8	1	1 with embryo
F	4	128/250	2	4	0	Seeds empty

[1]Test based on five samples.

stoppered flasks. No attempt was made to control the humidity as the object was to ascertain how long pollen would remain fully viable when kept in an ordinary refrigerator.

Periodic germination tests were first conducted on different pollen lots which were stored under these conditions from 1952 to 1955. The viability of all the lots gradually declined. Figure 12 illustrates the results of a germination test with two-year-old pollen. Viability of this pollen lot was very low and considerable desiccation was evident. Figures 13 and 14 illustrate the results of germination tests with two pollen lots which had been stored for one year. It can be seen that the viability of the one lot has declined much more rapidly than that of the other and this variation in vigour was observed in the pollen from several different trees.

It should be realized, however, that germination tests do not necessarily give any indication of the fertilizing potential of the pollen. Accordingly some of the pollen lots which had been stored for two years were used for controlled crosses, the cones being removed in the fall and the seed extracted and cleaned. The results given in table 2 indicate that, although all the pollen lots showed some germination, the fertilizing potential had been drastically reduced as only two seeds contained embryos. Some cones were also removed from each of the trees a few weeks after pollination and a few of the ovules were sectioned and stained. Sections of the micropyle of two of these ovules showed ungerminated pollen twenty-three and thirty-two days after pollination, whereas pollen usually germinates from seven to ten days after controlled crosses.

In a second test, the same two pollen lots were used as pollen parents for the same parent trees for two successive years. The results given in table 3 showed that,

TABLE 3. Results of pollination with fresh and one-year-old pollen.

Tree	Pollen parent	Year of pollin.	Pollen[1] germin.	No. of cones collected	No. of cleaned seeds	Germin.	Cutting test of ungermin. seed
F	H	1954	240/250	33	420	400	0
		1955	244/250	4	67	2	9 with embryos
J	C	1954	230/250	41	1,051	397/420	0
		1955	240/250	13	555	39	96 with embryos

[1]Test based on five samples.

whereas the viability of these pollen lots had apparently not been affected by storage, the fertilizing potential had been drastically reduced as there was a highly significant decrease in the amount of viable seed produced. Duffield (1954) has pointed out that pollen germination tests in the pines can not be considered a reliable index of fertilizing ability as they only show that a certain percentage of grains is able to cross a certain functional threshold. Germination tests can be of some value, however, in preventing the use of sterile or very weak pollen in controlled crosses. The results of these germination tests definitely indicate that, under ordinary refrigeration storage, Douglas fir pollen loses its fertility rather quickly and that improved storage facilities are necessary if pollen can not be used the same year of collection.

Literature Cited

CUMMING, W. C., and F. I. RIGHTER. 1948. Methods used to control pollination of pines in the Sierra Nevada of California. U. S. Dept. Agric. Circ. 792.

DUFFIELD, J. W. 1950. Techniques and possibilities for Douglas fir breeding. J. For. 48: 41-45.

————. 1954. Studies of extraction, storage and testing of pine pollen, Z. Forstgenet. 3: 39-45.

NIENSTAEDT, H., and H. B. KRIEBEL, 1955. Controlled pollination of eastern hemlock. For. Sci. 1: 115-120.

ORR-EWING, A. L. 1954. Inbreeding experiments with the Douglas fir. For. Chron. 30: 7-16.

Forestry and Wildlife Atlas For Czechoslokavia

Review by R. Stahelin
U. S. Forest Service, Washington, D. C.

The Central Research Institute of Czechoslovakia has published in 1955 a Forestry and Wildlife Atlas (*Lesnicky a Myslivecky Atlas*). It contains 120 size 8 in. × 12 in. sheets of well executed color maps. There are several detailed maps each of tree distribution, forest types, plantations of exotic trees, forest management, forest influences, and 12 maps on distribution of game fish, animals, and birds in Czechoslovakia. There are also forest type maps for all Soviet bloc countries except China. Of general interest are 25 plates containing 80 smaller maps showing the natural range of approximately 400 tree species of the northern temperate zone. One map shows the world distribution of antlered game. An enclosure gives translations of all titles and legends of maps in English and other languages. A 91-page text with many charts and maps furnishes a commentary to the atlas.

Effects of Temperature upon the Activity and the Behavior of the Douglas fir beetle

BY

J. A. RUDINSKY
J. P. VITE

THE DOUGLAS FIR BEETLE, *Dendroctonus pseudotsugae*, Hopk., is one of the most destructive insects in the Pacific Northwest, and we have long lacked basic information on this pest. Today the need for basic research is so well understood that there is no necessity to stress the connection between the behavior of destructive forest insects and their control.

The present study was made during the winter of 1955 and through April, 1956. The beetles used came from Douglas firs that were cut in the early spring of 1955 in the Oregon State College McDonald Forest and infested in late May. The infested logs, sealed by wax, were kept in the insectary until used in the laboratory. The dormancy of the beetles was broken by exposing the infested logs to 22°C. for various periods of time.

The external manifestations of life and the development and maturation of the Douglas fir beetle are influenced by temperature. The activity, however, is influenced directly by temperature only to a certain degree, whereas the development is always influenced directly. Moreover, a given temperature enables the insect to perform definite actions, the execution of which is then governed by the central nervous system and, in most cases, can be also regulated physiologically, by the senses. This regulation may depend to a large degree more on the outside temperature than on other climatic factors; it always lies, however, within the limits of a determined behavior typical of the condition and disposition of an individual. In other words, the rising outside temperature leads less to progressively increasing activity than to a decisive mode of activity, which is released by the central nervous system after the threshold temperatures are passed.

This phenomenon becomes especially clear in the flight. Although at 12°C. the Douglas fir beetle is able to fly, usually the beetle first starts to fly only about 20°C. But even at much higher temperatures the frequency of the wing beats scarcely changes. The boring activity also

---

J. A. Rudinsky is Associate Professor of Forest Entomology at Oregon State College, Corvallis, Oregon. J. P. Vité is Docent of Forest Entomology at Göttingen University, Germany, and was Fulbright Research Scholar in the United States, 1955-56. This paper contains part of the results of a cooperative research project sponsored by the Northwest Forest Pest Action Comm., the Forest Protection and Conservation Committee of the Oregon State Board of Forestry, the O. S. C. Forest Experiment Station, and the O. S. C. Department of Entomology. Acknowledgment is due to Dr. J. D. Lattin, O. S. C. Department of Entomology, R. L. Furniss and K. H. Wright, U. S. Dept. Agric., Forest Service, Pacific Northwest Forest and Range Experiment Station, for critical review of the manuscript.

can distinctly depend on the disposition of the beetle; thus, under certain conditions greater boring activity occurs in the lower temperatures than in the middle temperatures. On the other hand, the running beetle seems to utilize all its possible ability at a given temperature, under the supposition, certainly, that this temperature is an increase over the temperature at which the beetle was predisposed. But also here, beyond a clear threshold temperature, spontaneous central nervous reactions take place. And within the adaptation temperature or at decreasing temperatures, the running activity is governed by the central nervous system and by the senses.

The activity and the behavior of sexually mature beetles were investigated under different temperature conditions; the air humidity and the light conditions were taken into consideration wherever they had or were expected to have a substantial influence on the beetle activity. First of all, the limits of the beetle activity had to be established.

The Limits of Activity

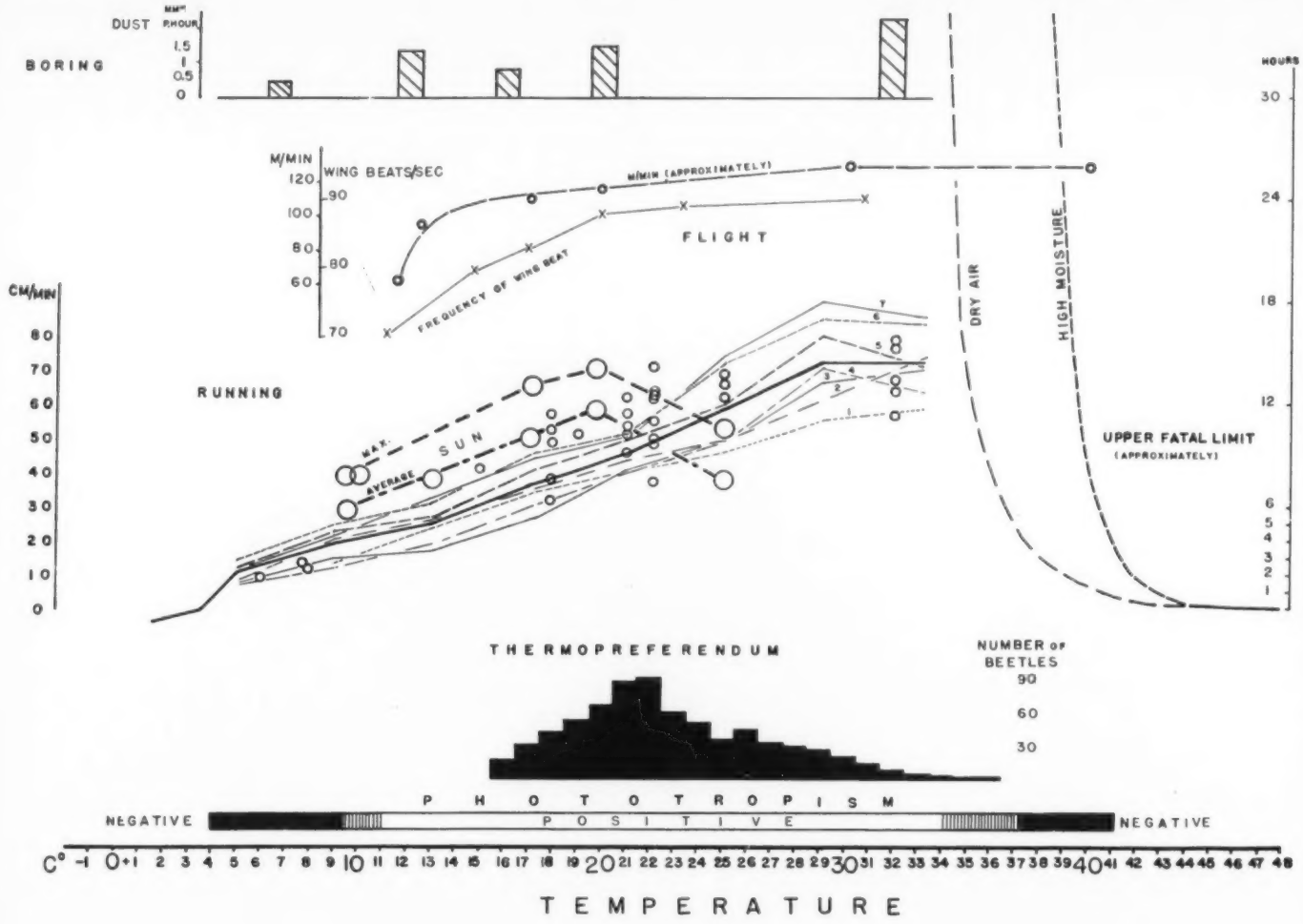
The absolute lower activity limit of the Douglas fir beetle lies between 0° and 1°C. Already at 1°C. the beetle reacts to contact or light stimulus with slow leg motion, and at 2°C. it is able to stand up but unable to move forward. Restrained though clearly regulated step movements take place at 3-4°C. At this temperature there is practically no boring activity, although some insignificant boring may take place. A few tenths of a degree below zero bring about a real "Kältestarre" (activity cessation due to low temperature), from which the beetle rouses relatively fast when influenced by even a small increase in temperature. For example, beetles which were kept for several days at a "Kältestarre" of −7°C. recovered in 16 minutes when exposed to + 22°C. They were able to perform activities characteristic for that temperature within 20 minutes.

To summarize, at low temperatures the Douglas fir beetle shows remarkably high activity. The zone of unorganized activity is narrow and low (1°C. to 3°C.), as is the zone between the "Kältestarre" and the beginning activity (about −0.5°C. to +0.5°C.).

A comparison is seen in *Ips typographus* L., a late-swarming beetle which shows some irregular movement only at 5-8°C. and is capable of weak, regulated activity only when the temperature reaches 10°C. (Vité, 1952).

The ability of the beetle to withstand heat is neither great nor unusually slight. Temperatures higher than 33-36°C. result in excessive activity; temperatures over 38-39°C., with low relative humidity and considerable exposure, cause heat injury. This injury becomes evident externally as exhaustion, or heat paralysis, followed by complete activity cessation and finally thermal death. These processes are the results of metabolic deficiency in breathing, the food supply, and particularly the water content of the insect. The occurrence of heat damage within this temperature zone depends upon the length of exposure, the relative humidity, and the nutrition. Nutrition, however, seems to play a limited role, because the higher temperatures prevent the regulated intake of food. Still higher temperatures may accelerate thermal death by breaking down the nervous system and the dependent vital functions. (See Weber, 1954).

Thermal death occurs between 47 and 49°C., independent of relative humidity; this temperature is lethal in 1-3 minutes. Temperatures between 45 and 46°C. can be withstood for several minutes (5 to 30), the time depending on the existing relative humidity as well as on the condition of the beetle. Below this temperature, however, the beetle shows considerable differences in thermal tolerance (Figure 1). At 40% relative humidity, the temperatures of 40-42°C. are lethal within a few hours, whereas the beetle can withstand the same temperatures at 100% relative humidity for days or at least for many hours. Since the subcortical relative humidity in the beetle galleries is close to the saturation point, one may assume that under natural


BORING
DUST MM³/HOUR
1.5
1
0.5
0
M/MIN
WING BEATS/SEC
120
90
100
80
80
60
70
M/MIN (APPROXIMATELY)
FREQUENCY OF WING BEAT
FLIGHT
CM/MIN
80
70
60
50
40
30
20
10
0
RUNNING
MAX.
AVERAGE
SUN
DRY AIR
HIGH MOISTURE
UPPER FATAL LIMIT
(APPROXIMATELY)
HOURS
30
24
18
12
6
5
4
3
2
1
THERMOPREFERENDUM
NUMBER OF BEETLES
90
60
30
PHOTOTROPISM
NEGATIVE
POSITIVE
NEGATIVE
C° -1 0 +1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28 29 30 31 32 33 34 35 36 37 38 39 40 41 42 43 44 45 46 47 48
TEMPERATURE

conditions only temperatures above 43°C. can be dangerous to the beetle.

Subcortical temperatures of this degree are by no means rare, particularly in the upper side of fallen trees exposed to direct sunrays (Graham, 1924). Spring attacks on such logs result in heavy brood mortality, and during the second or summer attack the beetles prefer the cooler and fresher sides of logs.

Boring Activity

The Douglas fir beetle is capable of gnawing at temperatures around 4°C.; at 15°C. it can bore into the bark within 5 hours, and at 25-28°C. within 40 minutes. The actual boring activity, however, depends to a great extent upon the disposition and the sex of the beetle. At higher temperatures, excessive activity or restlessness hinders the organized boring activity of the beetle.

In order to find the absolute boring capacity at different temperatures, beetles adapted to 20°C. were kept for 42-44 hours on fresh Douglas fir bark at different temperatures. At least 30 beetles were used in each experiment. The experiments were repeated three times with new beetles that had been adapted to 20°C. The result is shown in Table 1.

TABLE 1. Boring dust as related to temperature (extremes in brackets).

Average temperature	Average boring dust per beetle per hour in mm^3
32°C. (30°-34°C.)3	2.29 (1.82-2.70)
20°C. (18.5°-22°C.)	1.54 (1.02-2.43)
16°C. (15°-17°C.)	0.77 (0.62-0.97)
12°C. (11°-13°C.)	1.37 (0.95-2.18)
6.5°C.	0.42

With the exception of the results at 12°C., this experiment shows an expected increase in boring activity from a minimum in the zone of beginning activity at 6.5°C. to a maximum in the upper zone of organized activity. The conspicuously higher boring activity at 12°C. seems to result less from accident than from stimulating the beetles by cooling them from 20°C. to 11-13°C. That is, these temperatures lie at the limit where the positive phototropic behavior of the beetles becomes negative.

Another experiment was designed to ascertain the correlation between boring and temperature. For this purpose, a fresh section of log on which the bark had been smoothed was exposed for two days to newly emerged beetles. Once the beetles were inside, the log was hung over a wide funnel in such a way that the boring dust fell on the rim of a cardboard disk mounted upon the rotation cylinder of a hygrothermograph (Figure 2). After 9 days the boring dust was measured and plotted in two-hour intervals against the temperature curve shown by the hygrothermograph.

Independent of the general decrease of boring activity as the egg laying progresses, a rhythm corresponding to the daily changes of temperature is evident. This rhythm follows the changes in the outside temperature with a lag of about 6 hours because of the insulating effect of the bark. Thus, the greatest boring activity occurred in the late evening or midnight, and the least activity during the forenoon (Figure 3). The temperature of 17°C. resulted in a great decrease of boring dust, as did the temperature of 15°C., and on the 6th day a decrease of temperature below 10°C. for 14 hours reduced the boring activity correspondingly. Intensive boring-in lasted for 6 days before the boring activity lessened considerably, and after 7 days it decreased rapidly. The same can be said about a parallel experiment, which ran first under the same temperature conditions

FIGURE 1. *Beetle activity in relationship to temperature. The temperature Centrigrade is plotted on the horizontal axis. In the range of phototropism, the indifference reactions are shown in the lined field. On the vertical axes, running activity is given in centimeters per minute (cm /min), flight activity in meters per minute (m/min), wing beat frequency in wing beats per second, and boring activity in cubic millimeters of boring dust per beetle and per hour (mm^3/ hour).*

FIGURE 2. *Apparatus for collecting boring dust at different temperatures.*

but from the 8th day with a lower temperature. Here also one insignificant increase in boring dust corresponds to the increased temperature on the 9th day (Figure 3). The females began to deposit eggs on the 2nd day, and many males left the galleries between the 4th and 8th day. The considerable decrease in boring dust may have several causes: the completion of the entrance galleries, the males' departure and the closing of the entrance hole, and the decrease in boring activity of the females during the egg-laying period.

The results of these experiments give some understanding of the behavior of the Douglas fir beetle in its natural habitat. At moderate temperatures, over 10°C., the beetle has relatively high boring ability, the performance of which, however, depends on its condition and disposition. Warmer temperatures, between 22-32°C., enable the beetle to bore through even thick bark within a relatively short time. This relationship between boring activity and temperature is basically important to the beetles' success or failure in breaking down the resistance of standing trees, and therefore to the primary destructiveness of the Douglas fir beetle.

Flight

Temperature has little direct influence on the flight capacity of the Douglas fir beetle; however, it delimits the zone of general flight ability. Of greater importance, first of all, is the condition of the beetle, or its ability and disposition to fly or not. Generally, both sexes are ready to fly as soon as they emerge from their galleries. They attack whenever the preferred breeding material is present, which occurred in the experiment even without free flight. Under natural conditions the beetles need not fly over long distances. In an epidemiological sense, the flight capacity, velocity and duration, plays a subordinate yet integral role compared to the beetle behavior, the orientation during the flight, and the wind dispersal.

At air temperatures below 18°C., the wings are spread only rarely and then for purposes other than take off (for example, to help the beetle to stand). Between 18°C. and 20°C., flight attempts do not occur while there is no sun. Temperatures around 22°C. regularly cause the beetles to take

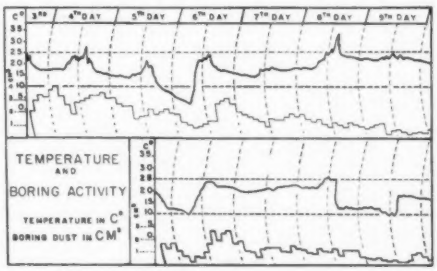


FIGURE 3. *Correlation between boring activity and temperature from the third day to the ninth day after attack and, in a parallel experiment, from the fifth day to the ninth day.*

off, and at higher temperatures, particularly around 25°C., spontaneous flight attempts are the rule. At those temperatures, the flight inclination is increased considerably by radiation, particularly that of sunshine; and at takeoff, the light source alone directs the beetle. In addition, at temperatures between 25° and 32°C., the flight seems to gain in flexibility. For example, after a collision with an object, the beetle is oftener able to continue the flight without falling than at lower temperatures. On the other hand, the flight velocity and the frequency of wing beat remain relatively constant.

The beetle flight was observed on a flight mill as modified by Chapman (1954) after Hocking (1953). For this purpose, a beetle was attached by a wax thread to a narrow cellophane strip that revolved on a needle. The velocity achieved in this way depended upon the radius of the rotating arm, and thus the air resistance against it, and the flight modus of the beetle. On this apparatus, one can make comparative studies of the flight and its duration under different temperature conditions but not of the velocity of the free-flying beetle.

Duration of flight. From the flight-mill studies, the following results were obtained. The beetle is capable of flying several hours daily for three days. The flight is often interrupted for various periods of time; continuous flights for ½-2 hours are frequent, and according to Chapman (1954, page 4), flights for 4 hours are "frequent and even flights of six hours are not uncommon". The same author recorded a flight maximum: "The maximum duration of continuous flight so far observed is over nine hours and that particular insect flew an additional six hours after an eight-hour rest and thus was in flight for 15 out of 23 hours".

Relative velocity of flight. Further experients on the flight mill showed that the Douglas fir beetle is able to perform long flights even at 17.5°C. At 12.5°C. the flight ability decreases markedly; slightly cooler, estimated at 11.5°C., the beetle was barely able to move the mill arm. Below this temperature, definitely at 10°C., flight stops completely. The average flight capacity of a beetle in the same position, for instance, was: at 40°C., 43.5 m/min. (meters per minute); at 31°C., 43.5 m/min; at 21°C., 39 m/min; at 17.5°C., 37.2 m/min; at 12.5°C., 32.5 m/min; and at 11°C. for a short time, 20.5 m/min. The influence of solar radiation is stimulating to such a degree that even at 21°C. the greatest flight performance is achieved, as is easily proved by moving the flying beetle from the shade into the sunshine.

Even if one considers a possible fault in the arrangement of the experiment, the differences in flight velocity obtained at different temperatures are still surprisingly small. If different temperatures caused even a moderate difference in flight activity, it would appear in the frequency of wing beats. Instead, this frequency —setting aside small individual differences— depends hardly on other factors than air pressure, and is therefore very constant. For that reason, the wing beat frequency was measured apart from the flight-mill experiment. To count the wing beats, the known method of a sooted cylinder was used. The side of a Petri dish was sooted over a candle and centered on the disk of a phonograph. A flying beetle, fastened on a cellophane strip, was brought so close to the side of the revolving Petri dish that only its wing tips during their downward beats touched the side (during the upward beat the wings are somewhat retracted). Each mark in the soot, then, represented one completed wing beat (Figure 4).

The values thus obtained show a pattern similar to the flight velocity. At 11°C. the wing beats average 71 per second, and at 15°C. they average 80. At 17°C., however, the number of wing beats is only

FIGURE 4. *The wing beat frequency of two beetles (upper and lower rows) as marked on a sooted Petri dish. The second line in each pair was marked when the Petri dish was revolved a second time in order to show the regularity of the wing beats.*

slightly higher. Then, at 20°C. the frequency reaches 88 and thereafter changes little, and even at 31°C. it increases only to 90 wing beats per second. In comparison to the flight velocity obtained on the flight mill, the wing beat frequency increases only gradually up to 20°C. (Figure 1). It is of special interest to note that the lower temperature limit which brings the optimal frequency of wing beats coincides with the temperature (18-20°C.) that enables the beetle to start flying.

Actual flight velocity. To learn the actual velocity achieved by the beetle in free flight, we attempted to get at least approximate data on the flight velocity in a windfree room. Here a distinction had to be made between the 'take off velocity" and the velocity of the "straight flight". The former was measured by stop watch from the time the beetle took off until it reached a light source 3 meters away; and the latter by stopwatch from the time the beetle crossed a marked line in straight flight until it reached the light source. In both cases, the figure given is minimum, because only forward flight, without small horizontal or vertical deviations, could be measured.

The starting velocities were about 95 m/min; the straight flight velocities of 50 newly emerged beetles lay consistently between 94 and 136 m/min with an average of 115 m/min. They amount to from double to triple the speed on the flight mill. The difference in the flight speeds during take off and later may result partly from the different positions of the body. While the beetle is rising, its body is in an almost vertical position, which it later changes (by stretching the abdomen) into a horizontal position, and thus it gains in velocity.

Considering the flight activity as a whole, one sees that the flight performance remains almost uniform within a relatively wide range of temperature. The threshold temperature which by necessity leads to a take off attempt is higher than the temperature necessary for continuing flight. In other words, the beetle does not take off at temperatures lower than 18-20 degrees, but once flying it is able to fly even though the temperature goes down to 13°C. This means that without doubt under natural conditions the beetle can fly through air masses cooler than the threshold temperature. The investigation of the factors in flight performance, duration and velocity, gives a basis for approximating the average flight capacity. Accordingly, it is apparent that the beetle can fly three days for at least four hours daily and so cover at least fifty miles in active flight—even when one disregards the enormous possibilities of transportation by air currents. This statement is made without overlooking the fact that the beetle's flight under natural conditions is most often only a search flight for suitable, proximate breeding material, and not a migratory flight. In such a search flight, the main role is played by the ability to direct the flight towards suitable breeding material. Apparently, this ability depends to a high degree upon temperature, and it seems to reach its optimum at the warmer temperatures.

Running Activity

The influence of temperature on the running activity of the beetle is evident and can be measured easily. Therefore, by comparing the different running performance and behavior at various temperatures, one can draw specific conclusions about the relationship between temperature and the activity of the beetle.

Because running activity depends on the double influence of temperature (on the process of muscle contraction and on the relaxation of muscles by the nervous system), it is necessary to insure that the beetles used in measuring running activity at different temperatures are stimulated in the same way. This was accomplished by exposing the beetles to low temperatures and then warming them up gradually to the desired temperature. The adaptation of the beetle to the new temperature takes place within 3 minutes. Because of individual differences it is best to use the same beetle in the series of tests with increasing temperatures. Nevertheless, the perform-

ance of a fresh beetle lay within the same narrow zone. If, however, the beetles are brought from warmer to lower temperatures, the stimulus for running is sharply decreased, and their performance is diminished.

The results of these experiments are presented in Figure 1. Each point corresponds to an average performance of five separately measured beetle runs (expressed in cm per minute), whether they belong to the main series of beetles (—broken or thin lines) or to supplementary measurements at single temperature degrees for which new beetles were always used (small circles). The heavy line represents the means of beetles 1-7, which were used in tests beginning with low temperatures and continuing to higher temperatures. Also, the procedure was reversed, and beetles exposed at first to higher temperatures were used in an experiment with decreasing temperatures. Their performance (mean of 10 beetles) was smaller, yet it lies at the lower limit of the range of the main experiment.

At 4°C. the beetle is capable of regulated running, and at 5°C. the velocity is around 10 cm/min. Thereafter, the velocity increases almost linearly to 29°C., where it averages 70-80 cm/min; there it may remain for a few degrees, but usually it decreases. This indicates that the optimal zone of organized activity (the optimum for regulated movements) is passed. The subsequent excessive activity (with its symptoms of circling, falling down, etc.), between 33° and 46°C., results in the disintegration of regulated movements, although some beetles may still occasionally show maximal performances.

The outside temperature is decisive in the activity of the beetle only when it equals the body temperature. Under natural conditions, however, the body temperature is often increased by radiation. Since it is difficult to measure the radiation energy absorbed by beetles, the running tests were performed in the sunshine. It was hoped thus to learn more about the increase in beetle activity caused by radiation. The tests were conducted at mid day (between 11:30 A.M. and 12:30 P.M. in the first week of March in Corvallis, Oregon). At an air temperature of 9°C., beetles placed in the sun where a thermometer recorded 16°C. showed activity typical of 16-17°C. in the shade (Figure 1, big circles). The increased activity is explained by the solar radiation. At an air temperature of 25°C. with the thermometer recording 42°C. in the sun, the beetles showed activity typical of 40-45°C. without radiation. Thus, at 17°C. air temperature (thermometer in the sun 33°C.) and at 19.5°C. (thermometer in the sun 35°C.), the beetles became particularly restless. At 25°C. air temperature, they showed excessive activity, and about one half of the beetles succumbed to thermal paralysis in a short time: an effect that occurs without solar radiation around 45° C. It is true that these tests had to be performed behind a window glass in the laboratory where the effect of sunrays was increased unnaturally. Nevertheless, these data give an impression of the radiation potential, which, for instance, can cause the beetles to fly at moderate temperatures and to avoid attacking the unfavorable, sun-exposed parts of logs.

Reactions

The reactions of the Douglas fir beetle resulting from temperature within its limits of activity may be summarized as follows:

	Degree of activity	Threshold temperature
I	Unorganized activity	over 0.5°C.
II	Limited activity	" 4°C.
III	Moderate activity and flight ability	" 10-11°C.
IV	Higher activity and take off ability	" 18-20°C.
V	Optimum of organized activity	" 26-28°C.
VI	Restlessness and thermal paralysis	" 33-36°C.

Temperature also influences the behavior of the beetle through its sense organs, particularly the light sense. When the temperature went below 11°C., the beetles began to change their phototropism from positive to negative, and at 9°C. all beetles in the test behaved negatively. Similarly, there is a change in the phototropic behavior of the beetle at higher temperatures, especially the higher radiation temperature,

which the beetle tries to avoid by running towards the shade. At 34°-37°C., the zone of indifferent behavior is relatively large, since it practically coincides with the zone of excessive activity; in the zones of strong light-shade contrasts, therefore, the negative phototropism is the only guidance noticeable at this state of unorganized activity.

Also, the temperature itself serves, to a limited extent, as a stimulus to orient the beetle in space. But the beetle has no distinct sense of temperature; rather its preference of a special temperature is governed by the central nervous system (von Buddenbrock, 1939). The beetles placed in a temperature-preference apparatus (so-called temperature organ) clearly avoided the temperatures above 32°C. and preferred temperatures below 27°C., particularly around 21 and 22°C.

Altogether 700 beetles were used in 9 experiments in the apparatus, which was heated at one end and cooled at the other, so that a gradient of temperature from 10° to 45°C. resulted inside (Figure 5). After being introduced, the beetles were allowed to settle for one hour at their preferred temperatures before the count was taken. From the 700 beetles, 304 preferred the temperatures between 20 and 23°C., 161 beetles gathered between 24 and 27°C., 90 between 28 and 32°C., and only 15 beetles over 32°C. On the other hand, the beetles which were found at the cool end of the trough were influenced too strongly by other factors to give objective data (compare Gunn and Walshe, 1942). These beetles did not return to the warmer end before their ability to move was reduced by the temperatures between 10° and 16°C. Also, the condensation in the cool end may have hindered their escape. Therefore, the beetles found below 16°C., as well as those which left the trough, were not included in the evaluation.

The temperature-preference apparatus was made as described by Nieschulz (1933) and used also by Vité (1952). A U-formed copper trough 48″ long, 3½″ wide, and 2″ high was imbedded in a layer of insulating fiber glass within a wooden container. Cross walls were welded in the trough at a distance of 8″ from each end, thus enclosing the experimental area and allowing one end to be heated by hot water and the other cooled by cold water. Fourteen thermometers were inserted in openings in the wooden container, so that they touched the bottom of the copper trough under the experimental area. The floor of the trough was covered with small bark pieces of the same size, upon which the beetles were uniformly distributed. The bark and beetles were covered by a cellophane strip, over which fiber glass was packed for insulation. Fresh bark pieces equalized the air humidity, provided equal contact possibilities for the thigmotactic behavior of the beetles, and allowed space for movement under the cellophane strip. The cellophane strip, together with the insulating fiber glass, eliminated the vertical differences in temperature. Phototropic reactions were avoided by covering the trough with a lightweight board.

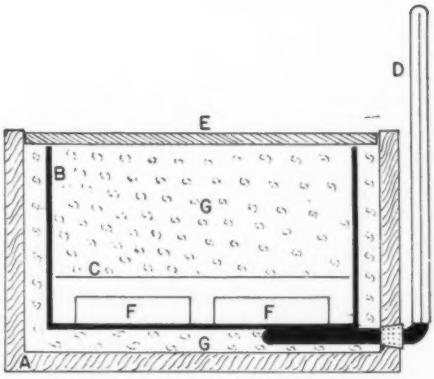


FIGURE 5. *Cross section of the temperature-preference apparatus. A. Wooden container. B. Copper trough. C. Cellophane strip. D. Thermometer. E. Lightweight board. F. Pieces of bark. G. Fiber glass.*

Summary

Dendroctonus pseudotsugae shows well-organized activity between 10° and 34°C. The beetle still has sufficient capability for boring in and for locomotion at the lower limit. This limit is also characterized by loss of the beetle's ability to fly and by the change from positive to negative phototropism — the latter change marking the

behavior of the beetle outside of the bark. At the upper limit, the shock reactions of restlessness and predominantly negative phototropic behavior lead the beetles to avoid higher temperatures than 33°C., which sooner or later would cause thermal paralysis and finally thermal death. The same limit restricts the regulated locomotion and the ability to bore in, whereas the flight performance remains the same.

Within this clearly defined zone of activity, there are two distinct temperature spans: the first, between 18 and 22°C., gives the condition to start flying; the second, between 26 and 32°C., encloses the optimum for regulated locomotion. It appears that the thermopreferendum of the Douglas fir beetle lies between these two spans of temperature.

Literature Cited

BUDDENBROCK, W. v. 1939. Grundriss der vergleichenden Physiologie. 2nd ed. Berlin.

CHAPMAN, J. A. 1954. Flight of *Dendroctonus pseudotsugae* in the laboratory. Bi-monthly Prog. Rept. Div. Forest Biol. Dpt. Agr. Can. 10:4.

GRAHAM, S. A. 1924. Temperature as a limiting factor in the life of subcortical insects. J. Econ. Ent. 17:377-383.

GUNN, D. L., and B. M. WALSHE. 1942. The biology and behaviour of *Ptinus tectus Boie.* (Coleoptera: Ptinidae), a pest of stored products. IV. Temperature preference .J. Exp. Biol. 19: 133-140.

HOCKING, B. 1953. The intrinsic range and speed of flight of insects. Trans. R. Ent. Soc. Lond. 104: 223-345.

NIESCHULZ, O. 1933. Ueber die Bestimmung der Vorzugstemperatur von Insekten (besonders von Fliegen und Muecken). Zool. Anz., 103: 21-29.

VITÉ, J. P. 1952. Temperaturversuche an *Ips typographus* L. Zool. Anz., 149: 195-206.

WEBER, H. 1954. Grundriss der Insektenkunde. 3rd ed. Stuttgart.

Nitrogen Loss From Granitic Soils on Cheatgrass Brome Range

BY
HAROLD F. HAUPT

THE STEEPLY SLOPING, granitic soil-grassland portion of the Boise River watershed in southwestern Idaho has long been recognized as a source of damaging sediment in the costly irrigation system of Boise Valley. A range erosion survey (Renner, 1936) showed south-facing slopes on which the native perennial grasses had been replaced by the annual cheatgrass brome (*Bromus tectorum*) to be eroding most seriously. Infiltrometer tests confirmed the survey findings and also showed that a single artificial rainfall removed substantial quantities of nitrogen from the surface soil (Craddock and Pearse, 1938). The nitrogen losses were believed to be a contributing factor to the slow recovery of a protective cover on the eroding lands.

In the more recent studies in the same locality Packer (1951a) found a 70-percent ground cover of plants and litter was necessary for minimizing soil erosion and storm runoff. He also found that still greater amounts of ground cover were necessary for maintaining soil stability when the surface soil was disturbed by trampling similar to that caused by heavy grazing (Packer, 1951b and 1953).

As an adjunct of the latter studies, tests were made to determine the amount of nitrogen losses from cheatgrass brome range sites during a single simulated major summer storm and the extent to which those losses were affected by trampling disturbance and amount of ground cover.

Plan of Study

Sixteen areas about 12 by 12 feet in size were located on south-facing cheatgrass brome slopes in the foothill portion of the Boise River watershed. The areas were chosen in pairs, each pair having nearly similar amounts of plant and litter cover. The 8 paired areas were chosen so as to include a range of ground cover from about 30 percent to more than 90 percent.

A 6- by 6-foot plot was centrally located in each of the paired areas. Soil sampling zones 3 by 6 feet in size were then established on three sides of each plot. The zones were located so that half of each was within the plot and the other half was outside.

At 3 random locations in each zone outside the plot, all plant litter not incorporated with the soil was removed and a steel ring 3.5 inches in diameter was driven into the soil to a depth of 0.5 inch. Soil was lifted from each ring with a spoon. The 3 ring samples were composited into a single sample, after the procedure followed by Cline (1945). This provided a total of 48 "before treatment" soil samples.

One study area in each pair was then randomly selected for trampling disturb-

The author is Forester, Intermountain Forest and Range Experiment Station, Forest Service, U. S. Department of Agriculture, Ogden, Utah.

TABLE 1. Total nitrogen content of surface one-half inch of soil before and after rainfall, nitrogen losses, overland flow, and soil eroded on untrampled and trampled cheatgrass brome plots.

	Plot and soil sample	Ground cover	Total nitrogen				Overland flow	Soil eroded
			Before	After	Losses			
		Percent	*Pounds per acre*	*Pounds per acre*	*Pounds per acre*	*Percent*	*Percent*	*Pounds per acre*
UNTRAMPLED	1-a		59.4	56.5	2.9			
	1-b		61.3	61.3	0.0			
	1-c		53.6	51.7	1.9			
	Avg.	33	58.1	56.5	1.6	2.8	19.4	2,499
	2-a		83.0	70.9	12.1			
	2-b		74.7	68.0	6.7			
	2-c		67.1	61.3	5.8			
	Avg.	32	74.9	66.7	8.2	10.9	14.1	1,066
	3-a		149.0	106.3	42.7			
	3-b		70.9	62.8	8.1			
	3-c		108.2	86.2	22.0			
	Avg.	52	109.4	85.1	24.3	22.2	10.0	858
	4-a		95.8	63.2	32.6			
	4-b		101.5	103.5	- 2.0			
	4-c		96.3	66.1	30.2			
	Avg.	64	97.9	77.6	20.3	20.7	4.6	97
	5-a		99.6	96.3	3.3			
	5-b		94.4	99.2	- 4.8			
	5-c		98.7	80.5	18.2			
	Avg.	72	97.6	92.0	5.6	5.7	1.5	21
	6-a		94.8	94.8	0.0			
	6-b		99.2	88.6	10.6			
	6-c		87.2	79.5	7.7			
	Avg.	70	93.7	87.6	6.1	6.5	2.0	10
	7-a		204.0	225.1	-21.1			
	7-b		192.6	196.4	- 3.8			
	7-c		160.0	164.8	- 4.8			
	Avg.	94	185.5	195.4	- 9.9	- 5.3	0.0	1
	8-a		130.3	127.4	2.9			
	8-b		129.3	145.6	-16.3			
	8-c		186.8	156.2	30.6			
	Avg.	94	148.8	143.1	5.7	3.8	7.1	55
	Subtotal average		108.2	100.5	7.7	[1] 7.1	7.3	576
TRAMPLED	9-a		95.3	72.8	22.5			
	9-b		86.3	73.8	12.5			
	9-c		78.6	68.0	10.6			
	Avg.	42	86.7	71.5	15.2	17.5	28.6	2,827
	10-a		81.4	51.7	29.7			
	10-b		57.5	35.4	22.1			
	10-c		81.4	58.9	22.5			
	Avg.	28	73.4	48.7	24.7	33.7	33.6	4.669
	11-a		82.4	57.5	24.9			
	11-b		91.9	54.6	39.3			
	11-c		74.2	69.0	5.2			
	Avg.	49	83.5	60.4	23.1	27.7	29.1	2,900
	12-a		107.8	107.3	0.5			
	12-b		88.1	95.8	- 7.7			
	12-c		124.5	103.5	21.0			
	Avg.	55	106.8	102.2	4.6	4.3	11.1	454
	13-a		84.3	89.1	- 4.8			
	13-b		123.6	113.0	10.6			
	13-c		103.9	112.1	- 8.2			
	Avg.	72	103.9	104.7	- 0.8	- 0.8	16.9	1,356
	14-a		166.7	136.0	30.7			
	14-b		160.9	141.8	19.1			
	14-c		201.2	158.4	42.8			
	Avg.	74	176.3	145.4	30.9	17.5	5.6	361
	15-a		196.4	168.6	27.8			
	15-b		128.8	133.2	- 4.4			
	15-c		227.0	160.0	67.0			
	Avg.	93	184.1	153.9	30.2	16.4	2.1	452
	16-a		140.3	133.2	7.1			
	16-b		121.7	136.0	-14.3			
	16-c		153.8	116.4	37.4			
	Avg.	85	138.6	128.5	10.1	7.3	1.3	265
	Subtotal average		119.2	101.9	17.3	[2]14.5	16.0	1,660
	Total average		113.7	101.2	12.5	[2]11.0	11.7	1,118

[1]Significant at 10-percent level.

[2]Significant at 1-percent level.

ance. Trampling was confined to the 6- by 6-foot plot within each area. Trampling was accomplished by striking 60 percent of the plot with a steel disk 0.2 square foot in area. This trampling closely simulated the disturbance observed on similar range that was heavily grazed by cattle.

By means of a type F infiltrometer, 1.8 inches of artificial rainfall in 30 minutes was then applied to each 6- by 6-foot plot. This amount and rate of rainfall is typical of a major summer storm in that locality (Packer, 1951a). Records were made of the amount of storm runoff and of the soil eroded.

"After storm" soil samples were then taken from the portions of the 3 sampling zones within each plot. The same procedure was used as for the "before" samples. The 48 "before" and 48 "after" samples were analyzed for total nitrogen by a modification of the standard Kjeldahl method (Assn. Off. Agric. Chem., 1945).

The difference between the amount of nitrogen found in the "before" and "after" samples was assumed to represent a nitrogen loss or gain. Similar comparisons were described by Utz, *et al.*, (1938) and were made by Craddock and Pearse (1938). The validity of this comparison is based on the general knowledge that 98 to 99 percent of the nitrogen in the soil is in an organic form, which precludes any significant translocation of nitrogen by leaching during a single rainstorm. Instead, if any appreciable quantity of nitrogen is lost, it must be removed as a constituent of the organic materials carried in suspension by surface runoff and raindrop splash.

Initial Nitrogen-Ground Cover Relations

Nitrogen in the surface soil before the plots were subjected to the artificial rainstorm averaged 113.7 pounds per acre for all plots. The average of 108.2 pounds per acre on the untrampled plots was not significantly different from the average of 119.2 pounds per acre on the trampled plots (Table 1).

The initial amounts of nitrogen in the individual soil samples varied from 53.6 to 227.0 pounds per acre. These variations were found to be directly and highly correlated with amounts of ground cover. On the average, sites having a ground cover of 30, 60, and 90 percent had approximately 70, 100, and 160 pounds of nitrogen per acre, respectively (Fig. 1).

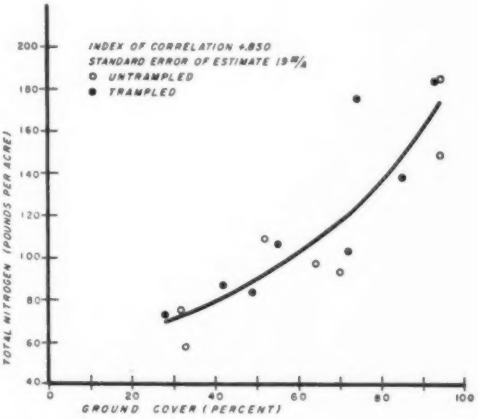


FIGURE 1. *Initial nitrogen content of surface one-half inch of soil in relation to amount of cheatgrass brome ground cover.*

Storm Effects

An average of 11.7 percent of the applied rainfall left the plots as overland flow. This runoff carried with it an average of 1,118 pounds per acre of eroded soil.

The nitrogen content of all soil samples taken after the artificial rainstorm averaged 101.2 pounds per acre. This is 12.5 pounds per acre and 11 percent less than the initial nitrogen content. This difference is highly significant statistically,[1] notwith-

[1]Loss value was subjected to a "t" test of differences between averages for paired observations.

standing the fact that in 11 out of 48 pairs of "before" and "after" soil samples there was a net nitrogen gain.

The average nitrogen loss obtained in this study is less than half the 25-percent loss measured by Craddock and Pearse in an earlier study (unpublished). However, the earlier study was conducted on very sparsely covered cheatgrass brome areas, whereas the present study involved areas having dense as well as sparse plant and litter cover. The 11-percent average loss is believed to be more nearly representative of the losses to be expected on the average cheatgrass brome range in this area.

Trampling Effects

On the untrampled plots, the single rainfall application produced 7.3 percent runoff and an average of 576 pounds per acre of eroded soil. The soil samples taken after the applied rainfall averaged 7.7 pounds per acre less nitrogen content than those taken before the rainfall application. This 7.1-percent nitrogen loss is significant at the 10-percent probability level.

Nitrogen loss from the trampled plots was 17.3 pounds per acre, or more than double the loss from untrampled plots. The disturbed plots also yielded more than twice as much runoff water and three times as much soil loss as the untrampled plots. The over-all effect of trampling disturbance was therefore that of greatly increasing the losses of soil and water as well as nitrogen.

Ground Cover Effects

On both the untrampled and trampled plots there was a highly significant inverse relationship between the amount of ground covered by plants and litter and the amount of storm runoff and erosion (Packer, 1953). Nitrogen losses on both untrampled and trampled plots also tended to decrease with increased ground cover (Fig. 2), but tests of the regression coefficients showed these relations not to be significant at the 5-percent level of confidence. In other words, ground cover in this study was less effective in influencing nitrogen losses than in controlling storm runoff and erosion.

Why ground cover was not more strongly related to nitrogen losses is not clear. It may be that the greater amounts of runoff and erosion on the poorly covered plots caused relatively low nitrogen losses because there was less nitrogen on those sites to be lost. Conversely, nitrogen losses may have been low from the well-covered sites where the initial nitrogen content was high simply because those sites produced little runoff and lost little soil.

The tendency for fertility losses not to vary proportionately with amount of runoff and soil loss has been observed in other studies on agricultural lands. For example, in a study of Dunkirk soils, Bryant and Slater (1948) found that fallow sites lost 43 percent more nitrogen than corn plots in one storm, but lost seven times more soil. During another period of the year, the corn plots lost 3 percent more nitrogen than the fallow plots though the latter lost about five times more soil. Other workers (Massey

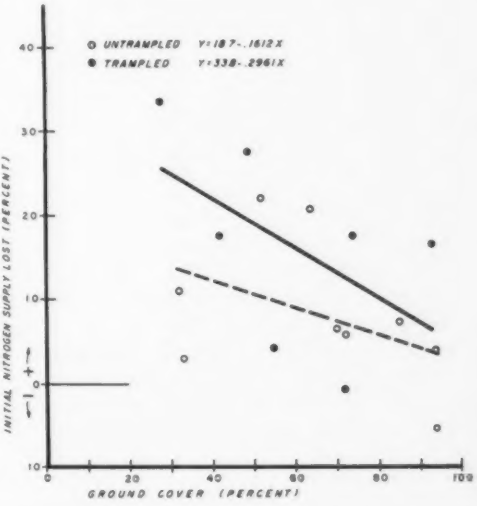


FIGURE 2. *Relation of nitrogen loss from surface 0.5 inch of soil to amount of plant and litter cover on cheatgrass brome plots.*

and Jackson, 1952; Rodgers, 1941; Slater and Carleton, 1942) have reported similar inconsistencies.

Summary

The nitrogen content of the surface 0.5 inch of soil on cheatgrass brome range sites in the Boise River watershed of south Idaho was found to vary directly with amount of plant and litter cover. A single application of artificial rainfall comparable to that in a major summer rain reduced the nitrogen content of the surface soil by an average of 11.0 percent. Artificial soil trampling similar to that caused by heavy grazing of cattle prior to artificial rainfall brought about nitrogen losses that were more than double thoese on undisturbed sites, and caused twice as much runoff and three times as much soil erosion. Runoff and soil losses were found to be highly correlated, inversely, with amount of plant and litter cover.

Nitrogen losses also tended to decrease with increased plant cover, though this relationship was not so clearly defined as that between ground cover and runoff and erosion.

Since the nitrogen content of the surface soil on poorly covered sites is inherently low and these sites are the most susceptible to storm runoff and erosion, any nitrogen loss from them further diminishes the chances for the development of a thicker plant cover.

Literature Cited

Association of Official Agricultural Chemists. 1945. Official and tentative methods of analysis. 6th Edition. Washington, D. C.

Bryant, J. C., and C. S. Slater. 1948. Runoff water as an agent in the loss of soluble materials from certain soils. J. Sci. (Iowa State College, Ames, Iowa) 22: 269-312.

Cline, M. G. 1945. Principles of soil sampling. Soil Sci. 58: 275-278.

Craddock, G. W., and C. K. Pearse. 1938. Surface runoff and erosion on granitic mountain soils of Idaho as influenced by range cover, soil disturbance, slope, and precipitation intensity. U. S. Dept. Agric. Cir. 482, 24 pp.

Massey, H. F., and M. L. Jackson. 1952. Selective erosion of soil fertility constituents. Proc. Soil Sci. Soc. Amer. 16: 353-356.

Packer, P. E. 1951a. An approach to watershed protection criteria. J. For. 49(9): 639-644.

———. 1951b. Status of research on watershed protection requirements for granitic mountain soils in southwestern Idaho. Res. Pap. No. 27, Intermt. For. Range Exp. Sta., U. S. For. Serv.

———. 1953. Effects of trampling disturbance on watershed condition, runoff, and erosion. J. For. 51: 28-31.

Renner, F. G. 1936. Conditions influencing erosion on the Boise River watershed. U. S. Dept. Agric. Tech. Bull. 528, 32 pp., illus.

Rodgers, H. T. 1941. Plant nutrient losses by erosion from corn, wheat, clover rotation on Dunmore silt loam. Proc. Soil Sci. Soc. Amer. 6: 243-246.

Slater, C. S., and E. A. Carleton. 1942. Variability of eroded material. J. Agric. Res. 65: 209-219.

Utz, E. J., Charles E. Kellogg, E. H. Reed, J. H. Stallings, and E. N. Munns. 1938. The problem: the nation as a whole. U. S. Dept. Agric. Yearbook, "Soils and Men." 84-110.

The Anthocyanin and Leuco-anthocyanins of Sugar Pine Seedlings

BY
STANLEY KRUGMAN

DURING THE GERMINATION and seedling stage of many pines a red coloration of the hypocotyl and cotyledons is present. Among the pines which have been reported to show this phenonemon are ponderosa pine (*Pinus ponderosa* Laws.), red pine (*Pinus resinosa Ait.*), and pinyon pine (*Pinus edulis* Engelm.) (Bates, 1925). It has been suggested that the red pigmentation serves as a protective device from excessive solar radiation, since the red pigmented parts of the plant absorb less of the long heat producing waves of the sun (Harvey, 1923).

Although coloration was noted by these investigators, the identity of the pigment or pigments responsible for the color was not determined.

This same pigmentation has been noted in sugar pine (*Pinus lambertiana* Dougl.) during and for a short while after germination. The duration of the pigmentation depends upon the temperature of growth. At 10°C, for example, the pigmentation may last from 30 to 70 days, but at 25°C it may be present for as little as one day. The coloration first appears in the hypocotyl and later in the cotyledons.

A preliminary exploratory study indicated that the red pigment was one of the anthocyanins. It also indicated that a colorless leuco-anthocyanin was also present.

It is the purpose of this study (1) to identify the anthocyanin and leuco-anthocyanin pigments in the seed, in the germinating embryo and in the young seedling, and (2) to establish the sequence of appearance and disappearance of the pigments in the germinating plant.

Chemistry of the Anthocyanins and Leuco-Anthocyanins

The anthocyanins are present in plant tissue as glycosides. Upon acid hydrolysis they yield one or more sugars and a sugar-free residue, the aglycone (anthocyanidin). Only seven aglycones have been found in plants (Bate-Smith, 1950), and of these, cyanidin, delphinidin and malvidin have been identified in the sugar pine seedling (Figure 1). At least 58 anthocyanins have been reported (Fulcher, 1954); but only one, malvin, was found in this study.

The leuco-anthocyanins are colorless, water soluble pigments whose structures have not been definitely established, although upon strong acid hydrolysis the leuco-anthocyanins will yield an aglycone (Rosenheim, 1920). The leuco-anthocyanins can best be characterized by the aglycone which they yield upon acid hydrolysis.

The author is at the School of Forestry, University of California, Berkeley. The article is revised from a dissertation submitted in partial fulfillment of the requirements for the degree of Master of Science in Forestry.

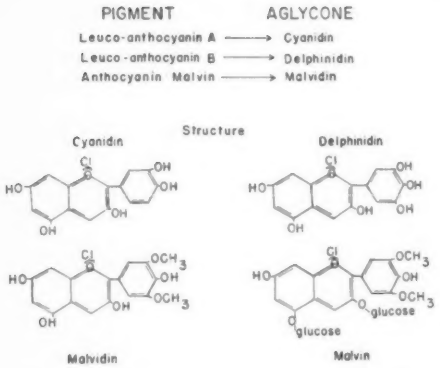


FIGURE 1. *The structures of the anthocyanin and aglycones identified in the sugar pine seedling.*

Anthocyanins have been known for a long time. As early as 1664 Robert Boyle reported on the behavior of this pigment toward acids and bases. Similar investigations were reported by Wray (1670), Lister (1671), Grew (1682), and others.

The presence of anthocyanins in seedlings is not uncommon. Pack (1921) found them in several species of Juniperus seedlings. Karstens (1939) detected anthocyanins in newly germinated plants of the common buckwheat (*Fagopyrum esculentum* Moench); Frey-Wyssling and Blank (1941) reported it in red radishes (*Raphanus sativas* L.) and red cabbage (*Brassica oleracea* var. *capitata rubra* L.); Gimesi et al. (1952) noted the presence of anthocyanins in the newly germinated plants of Love-Lies-Bleeding (*Amaranthus caudatus* L.); Favarger (1954) detected it in several species of *Saxifraga*.

In the genus *Pinus* anthocyanins have been identified but not studied in detail. Gertz (1906) reported their presence in the cones of pine but no identification was made. Robinson and Robinson (1934) found the anthocyanin, cyanidin-3-monoside, in the young cones of the Himalayan pine (*Pinus excelsa* Wallich) which they obtained from Sweden. Hayashi and Abe (1953) have detected and identified the anthocyanin, chrysanthemin, from the reddish purple staminate cones of the Japanese black pine (*Pinus Thumbergü* Parl.).

More widely distributed than the anthocyanins are the colorless leuco-anthocyanins postulated as precursors of the anthocyanins within the plant (Gortner, *et al.* 1949). The first report of the presence of a leuco-anthocyanin in the wood of pine was made by Robinson and Robinson (1933).[1] They also detected an aglycone cyanidin, from the hydrolysis of a leuco-anthocyanin found in the cones of Scotch pine (*Pinus sylvestris* L.), (Robinson and Robinson, 1934). Recently other cyanidin-yielding leuco-anthocyanins have been identified in the wood of maritime pine (*Pinus pinaster* Ait.), (Bate-Smith, 1954),[2] and in Monterey pine (*Pinus radiata*), (Hillis, 1954).

For a general review of the anthocyanin pigments the reader is referred to the first detailed record of this topic by Onslow (1925), and to more recent reviews by Blank (1947), and Paech (1955).

Materials and Method

Source of plant material and treatment. Sugar pine seed used in this study were collected at Pino Grande, California, during the fall of 1953. All seeds were from the same tree but not necessarily from the same pollen parent.

The seeds were first stratified in moist vermiculite at 5°C for 90 days and then germinated without the seed coat on sterile two percent water-agar in the dark at 10°C (Stone and Duffield, 1950).

Identification of the pigment. The paper chromatographic methods suggested by Bate-Smith (1948) and Cramer (1954) for the identification of anthocyanins were followed; however, certain modifications and additions were necessary.

The anthocyanin was identified by first identifying the aglycone and then in turn its related anthocyanin. The R_F values obtained were used only as rough guides

[1]Species was not given by the authors.

[2]The author listed this species as *Pinus maritima*.

TABLE 1. R_F values for the anthocyanin and aglycones identified in sugar pine seedlings.[1]

Solvent	Aglycone			Anthocyanin
	Cyanidin[2]	Delphinidin	Malvidin	Malvin
n-butanol 2N HC1	0.71(.68-.73)	0.36(.34-.37)	0.54(.50-.56)	0.82(.81-83)
m-cresol acetic-acid-water	.70(.68-.72)	.52(.50-.53)	.90(.87-.92)	.55(.54-.57)
n-butanol acetic acid-water (4:1:5)	.69(.68-.71)	.37(.35-.38)	.54(.53-.56)	.23(.20-.25)
n-butanol acetic acid-water (3:2:3)	.64(.59-.68)	.40(.37-.44)	.50(.45-.57)	.20(.18-.83)
acetic acid HC1 water	.50(.48-.51)	.31(.30-.33)	.60(.58-.61)	.82(.81-.83)

[1] R_F values were for chromatographs run at 21°C.
[2] The single value is the average for six runs, while the values in the parenthesis is the range.

(Table 1). Reference pigments and color reactions to various sprays were used for the positive identification of the unknown pigments (Hayashi and Abe, 1952).

Identification of the aglycone. (a) *Extraction.* Two to three grams of crushed pigmented plant material were hydrolyzed by boiling in 2N HC1 for 25 minutes. The aqueous solution was then filtered into a small test tube and to this was added 0.5cc of iso-amyl alcohol. After shaking, the pigment appears in the upper iso-amyl alcohol layer.

(b) *Separation.* The chromatograms were made on 7x20 inch Whatman No. 1 filter paper. The solution was concentrated on the starting line by spotting successive small amounts of the extract with a micropipette and drying with a current of warm air. The applications were repeated until sufficient pigment had been added to ensure a color on the developed chromatogram.

A number of solvents were tried. In general, the alcohol solvents caused fading of the spot unless a low pH was maintained during the separation (Bate-Smith, 1954). The solvents finally selected were: n-butanol-2N HC1 (1:1(v/v)); m-cresol-acetic acid-water (25:1:4(v/v)), (Bate-Smith, 1950); and water-acetic acid-concentrated HC1 (10:30:3(v/v)), (Bate-Smith, 1954).

Descending one and two-dimensional chromatography were tried. No improvement in the pigment separation was obtained by the use of two-dimensional chromatography in lieu of one-dimensional chromatography. Therefore, one-dimensional chromatography was used.

Each chromatogram consisted of a single sheet on which the spotting of the pigment was duplicated. Markers of known identity were placed on the same chromatogram as the unknown and all chromatograms were replicated.

The chromatograms were run in glass jars at a temperature of 21°±1C. Before each run the chamber was saturated with the vapors of the solvent for at least 24 hours. The paper was spotted with the pigment and equilibrated for 12 hours with the vapors of the solvent before irrigation. The time of irrigation varied between six and ten hours.

(c) *Sources of known aglycones.* The known aglycones used for markers in the identification of the unknowns were from natural sources known to contain these pigments. The same method of extraction and separation of the known pigments was used as with the unknowns described earlier. Table 2 shows the sources of the natural extracts used in this study.

Identification of the anthocyanin. Once the

aglycone is known, the anthocyanin can be quickly identified by chromatographing the unknown with known anthocyanins containing the aglycone previously identified.

(a) *Extraction.* Two to three grams of crushed pigmented plant material were heated gently for 25 minutes in a one percent HC1-ethanol (concentrated HC1-95 percent ethanol) solution. The resulting colored solution was filtered. From this the yellow, water soluble anthoxanthins were extracted by shaking with ethyl acetate (Robinson and Robinson, 1933). The ethyl acetate layer was then discarded. The ethyl acetate removed most, but not all of the anthoxanthins present.

(b) *Separation.* The chromatographic identification of the anthocyanin was similar to that discussed earlier for the aglycone. For this separation two additional solvents were used. These were n-butanol-acetic acid-water (4:1:5(v/v)), (Bate-Smith, 1948) and a modification of the last solvent, n-butanol-acetic acid-water (3:2:3-(v/v)).

(c) *Source of known anthocyanins.* Known samples of the anthocyanins used in this study were obtained from K. Hayashi, National Institute of Genetics, Japan. The peonin was originally extracted from the flower of *Paeonia albiflora* Pallas, and the malvin from the flower of *Rhododendron reliculatum* D. Don.

Identification of the leuco-anthocyanin. When both the leuco-anthocyanins and the anthocyanins are present in the same plant tissue, the problem of individual pigment identification is difficult since both the anthocyanin and the leuco-anthocyanin yield aglycones which may be identical after hydrolysis with 2 N HC1. In this study, however, this difficulty was avoided since the leuco-anthocyanins were identified in the newly germinated seedling before the anthocyanins had developed and also in the older seedling in which the red pigmentation was no longer apparent.

The extraction, separation, and identification of the leuco-anthocyanins were the same as discussed for the aglycones.

TABLE 2. Natural sources of aglycones used in this study.

Aglycone	Plant	Plant Part
Cyanidin	*Rhododendron Thomsonii* Hook.	Flower[1]
	Fagus sylvatica L.	Leaves[1]
Delphinidin	*Rhododendron rhodora* Don.	Flower[1]
Pelargonidin	*Pelargonium cucullatum* Ait.	Flower[1]
Peonidin	*Paeonia trolloides* Stafe x Stern	Flower[2]
Malvidin	*Rhododendron Californicum* Hook.	Flower[1]
	Rhododendron Augustinii Hemsl.	Flower[1]
	Fuchsia triphylla L.	Leaves[1]

[1]Lawrence, W.J.C. et al., 1939. The distribution of anthocyanins in flowers, fruit and leaves. Phil. Trans. Roy. Soc. of London. 230:149-178.

[2]Price, J.R. and V.C. Sturgess. 1938. A survey of anthocyanins. Biochem. Jour. 32(9):1658-1660.

Results

The anthocyanin and leuco-anthocyanins of the seed. Since the anthocyanin had been noted in the very young seedling, the question arose as to whether or not this pigment was in the need. The presence of leuco-anthocyanin also was investigated.

An examination of the endosperm tissue indicated that both pigments were absent. On the other hand, leuco-anthocyanins which yielded cyanidin and delphinidin upon strong acid hydrolysis were found in the embryo of the ungerminated seed. When the hypocotyl and the cotyledons of the embryo were tested individually, the two leuco-anthocyanins previously identified were found in both tissues. This is shown in chromatogram 1 of Figure 2; spots D and D′ are cyanidin and E and E′ are delphinidin. The lowermost row of spots, G, are anthoxanthin pigments not removed by the ethyl acetate washing. Spot G′ is also probably one of the anthoxanthins.

The anthocyanin and leuco-anthocyanins of the germinating embryo. Shortly after germination the embryo, with endosperm still attached, did not show any anthocyanin development; but, one to ten days later, anthocyanin pigmentation was apparent in the hypocotyl but not in the cotyledons (Figure 3, part 2). During this period chromatographic separation of the pigments extracted with strong acid yielded three spots with the hypocotyl extract and only two spots with cotyledon extract. The

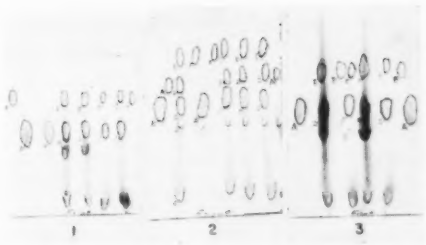


FIGURE 2. *Chromatograms of extracts of sugar pine seed and seedlings illustrating anthocyanin and leuco-anthocyanin pigment separation in the HCl-Acetic acid-Water solvent. Spots A, B, and F were extracted from known sources of aglycones; C, D, and E from the cotyledons; C′, D′, and E′ from the hypocotyl. Spots A, D, and D′ are cyanidin; F, E, and E′ are delphinidin; B, C, and C′ are malvidin; and G is an anthoxanthin. Chromatogram 1 is from an extract of an ungerminated embryo.*
Chromatogram 2 is from an extract of a ten day old germinated seedling.
Chromatogram 3 is from an extract of a 60-day-old seedling.

aglycones now detected were the result of the acid hydrolysis of the anthocyanin and leuco-anthocyanins present in the tissue. The aglycones found in the hypocotyl were identified as cyanidin, delphinidin, and malvidin. Those in the cotyledons were identified as cyanidin and delphinidin which are the same as those found in the embryo of the ungerminated seed. It would thus appear that malvidin is the aglycone associated with the red coloration of the hypocotyl.

Five days after this color first appears in the hypocotyl it becomes more intense and is present in the basal portion of the cotyledons as well (Figure 3, part 3). The pigments present at this stage of germination are shown in chromatogram 2 of Figure 2; spots D and D′ are cyanidin, E and E′ are delphinidin and C and C′ are malvidin. In succeeding days the red anthocyanin is found throughout the cotyledons (Figure 3, part 4, and Figure 4, parts 5 and 6). It was visually apparent that as the embryo matured the amount of anthocyanin in the hypocotyl decreased while the amount in the cotyledons increased. The red pigment was identified as malvin (Figure 5) and is a 3,5-diglucoside of malvidin.

The anthocyanin and leuco-anthocyanins of the newly germinated seedling. Between the thirtieth and forty-fifth day of germination the endosperm is pushed off by the cotyledons. Thus, with the removal of the endosperm the embryo becomes independent of the gametophytic tissue and the developing plant has reached the seedling stage. Extraction and chromatographic separation of the pigments revealed that the distribution of the anthocyanin and leuco-anthocyanins remain unchanged.

As the seedling matures, red pigmentation is still present but becomes less and less prominent in the cotyledons disappearing at a seedling stage of about 60 to 70 days (Figure 4, part 8). Chromatograms of the acid extract of the plant material following the disappearance of the anthocyanin pigment revealed the presence of leuco-anthocyanins which yielded cyanidin and delphinidin, but not malvidin (Figure 2, chromatogram 3).

Further extractions and chromatographic

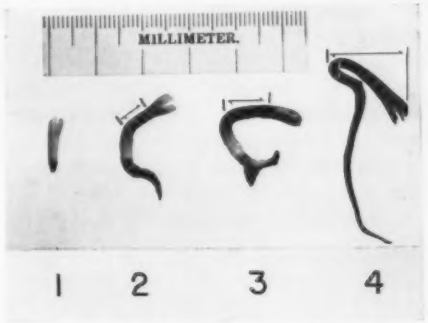


FIGURE 3. *Anthocyanin pigment development in the sugar pine embryo. The endosperm was removed to show the pigmentation. The brackets indicate the anthocyanin pigment during various stages of growth. (1). An ungerminated embryo from a 90-day stratified seed. (2). An 8-day germinated embryo. (3). A 12-day germinated embryo. (4) A 25-day germinated embryo.*

FIGURE 4. *Anthocyanin pigment development in the sugar pine embryo and seedling. The brackets indicate the anthocyanin pigment during various stages of growth. (5). A 30-day germinated embryo with the endosperm removed. (6). A 40-day germinated embryo with the endosperm removed. (7). A 50-day-old seedling. (8). A 70-day-old seedling.*

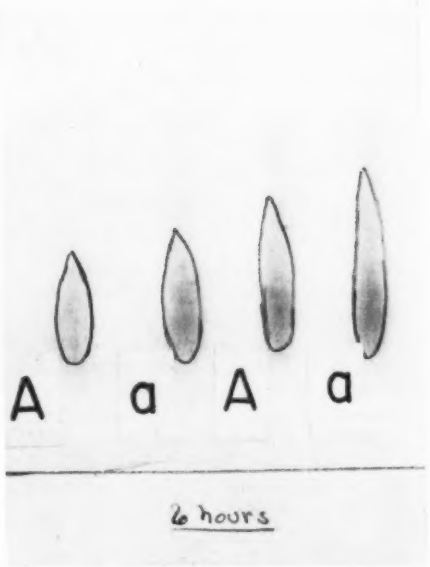


FIGURE 5. *Chromatogram illustrating the separation and identification of the anthocyanin in sugar pine seedlings. "A" is the spot from a sugar pine seedling extract. "a" is a spot from the flower extract of* Rhododendron reliculatum *known to contain the anthocyanin, malvin.*

separations of the leuco-anthocyanins were made of seedlings up to 150 days old. The results were the same as with the 60 to 70 day old seedlings. Only cyanidin and delphinidin could be detected.

Discussion and Conclusions

The presence of both anthocyanin and leuco-anthocyanin pigments in the same plant tissues plus the conversion of the leuco-anthocyanin into an aglycone in the *in vitro* system has suggested to some workers a possible relationship between the two groups of pigments (Simmonds, 1954, Hillis, 1955, Alston and Hagen, 1955).

Alston and Hagen (1955) found pelargonidin in the petals and peonidin in the sepals of the red flowers of *Impatiens balsomina*, while in the colorless bud they found leuco-anthocyanins which yielded cyanidin, delphinidin and pelargonidin. From this they concluded that those leuco-anthocyanins which do not correspond to the ultimate pigments disappear as the anthocyanins develop. It was concluded that the leuco-anthocyanins and anthocyanins are controlled by the same gene.

Similar results have been reported by Simmonds (1954) on the maturing bracts of the banana infloresence. He found leuco-anthocyanins in the young banana bract which yielded cyanidin and delphinidin on acid hydrolysis, while the mature bracts contain glycosides of peonidin, malvidin, cyanidin, and delphinidin. He was unable to determine the precise relation between the anthocyanins and leuco-anthocyanins

because upon acid hydrolysis both the anthocyanins yield aglycones which may be identical. However, by comparing the relative intensities of the anthocyanin and aglycone spots, he found that the leuco-anthocyanins had almost disappeared from the mature pigmented bract.

Both Alston and Hagen (1955) and Simmonds (1954) noted the presence of anthocyanins in the mature plant tissue; but they did not find leuco-anthocyanins which could serve as precursors of the anthocyanins since the leuco-anthocyanins yielded aglycones which were different from the ones found in the anthocyanins. Alston and Hagen detected glycosides of malvidin in the violet-purple flower of *Impatiens balsomina*, while in the unpigmented bud of the same plant they identified leuco-anthocyanins which yielded cyanidin and delphinidin. Both leuco-anthocyanins were absent from the mature pigmented flower. As noted earlier, Simmonds reported the presence of glycosides of peonidin and malvidin in the pigmented bracts, while the colorless bracts contained leuco-anthocyanins which yielded cyanidin and delphinidin.

In the sugar pine seedlings studied here the results were the same as the preceding ones. Leuco-anthocyanins from the young pine plants which would yield an aglycone associated with the anthocyanin, malvin, were absent.

Although the amounts of anthosyanin and leuco-anthocyanin were not followed, visual observations indicate that beginning with complete lack of anthocyanin pigmentation in the newly germinated embryo, there is a gradual build-up and then a reduction of the pigment as the plant matures until eventually the anthocyanin is once again absent. During the same period the leuco-anthocyanins appear to show no such change since they were detected in all stages of development. Were the leuco-anthocyanins precursors of the anthocyanins, one might expect them to disappear concomitantly with anthocyanin synthesis. The lack of this reciprocal relationship and the fact that the leuco-anthocyanins yielded aglycones different from that of the anthocyanins suggests that the leuco-anthocyanins are not direct precursors of the anthocyanins and that the two pigment groups are synthesized independently.

Summary

The anthocyanin and the leuco-anthocyanins in the ungerminated embryo and in the newly germinated seedling of sugar pines were identified chromatographically. The anthocyanin was found to be malvin and the leuco-anthocyanins yielded cyanidin and delphinidin.

From the sequence of anthocyanin and leuco-anthocyanin development in the germinating seedling plus the lack of a leuco-anthocyanin which could yield an aglycone corresponding to the anthocyanin malvin it is suggested that the leuco-anthocyanins are not direct precursors of the anthocyanins.

Neither the presence nor the distribution of anthocyanin is stable in the developing seedling. The pigment is first apparent in the hypocotyl; as the plant matures there appears to be a shift of the pigment to the cotyledons. By the time the plant is 30 to 70 days old the anthocyanin also disappears from the cotyledons.

Literature Cited

Alston, R. E. and C. W. Hagen. 1955. Relation of leuco-anthocyanins to anthocyanin synthesis. Nature 175:990.

Bate-Smith, E. C. 1948. Paper chromatography of anthocyanins and related substances in petal extracts. Nature 161: 835-838.

———— 1950. Title 6. Anthocyanins, flavones and other phenolic compounds. Symp. biochem. Soc. 3:62-73.

———— 1954. Leuco-anthocyanins 1. Detection and identification of anthocyanins formed from leuco-anthocyanins in plant tissue. Biochem. J. 58:122-125.

BATES, C. G. 1925. A key to the identification of some coniferous seedlings. J. For. 23:278-281.

BLANK, F. 1947. The anthocyanin pigments of plants. Bot. Rev. 13:241-317.

BOYLE, R. 1664. Experiments and considerations touching colours. London.

CRAMER, F. 1954. Paper chromatography, 2nd ed. St. Martin's Press, New York.

FAVARGER, P. C. 1954. Sur une fonction curieuse de l'albumen pendant la germination. Ber. Schweiz. Bot. Ges. 64:84-94.

FREY-WYSSLING, A. and F. BLANK. 1941. Physiological conditions of anthocyanins in etiolated seedlings. Nature 147:148.

FULCHER, M. J. 1954. A selected bibliography in pigments excluding chlorophyll. Master's thesis, Cath. Univ. Am.

GERTZ, O. 1906. Studier ofver anthocyan. Akademisk Afhandling, Lund. 97. 410 pp.

GIMESI, N., A. GARAY, C. FARKAS, and B. POZSAR. 1952. Physiology of pigment formation in plants. Acta. Biol. Acad. Sci. Hung. 3:247-259.

GORTNER, R. A., R. A. GORTNER, JR., and W. A. GORTNER. 1949. Outline of biochemistry. 3rd ed. John Wiley and Sons, Inc., New York, 1078 pp.

GREW, N. 1682. The anatomy of plants, with an idea of a philosophical history of plants and several other lectures, read before the Royal Society, London.

HARVEY, R. B. 1923. Red as a protective color in plants. Ann. Rep. Minn. State Hort. Soc. p. 279.

HAYASHI, K. and Y. ABE. 1952. Fundamentals on experimental procedures for the paper-chromatographic survey of naturally occurring anthocyan. Misc. Repts. Research Inst. Nat. Resourses (Japan) 28:1-11.

———— 1953. Studien uber anthocyane, XIII. Paper chromatographische ubersicht der anthocyane im pflanzenreich 1. Misc. Repts. Research Inst. Nat. Resources (Japan). 29:1-8.

HILLIS, W. E. 1954. The precursors of the red color in leather. Preliminary survey. J. Soc. Leather Trades Chem. 38: 91-102.

———— 1955. Formation of leucoanthocyanins in Eucalypt tissues. Nature. 175:597-598.

KARSTENS, W. K. H. 1939. Anthocyanin and anthocyanin formation in seedlings of *Fagopyrum esculentum*. Moench. Rec. Trav. Bot. Neerl. 36:85-179.

LISTER, M. 1671. Some observations, touching colours, in order to the increase of dyes and the fixation of colours. Phil. Trans. Royl Soc. of London. 6:2132.

ONSLOW, M. W. 1925. The anthocyanin pigments of plants. 2nd ed. University Press, Cambridge. 314 pp.

PACK, D. A. 1921. After ripening and germination of *Juniperus* seeds. Bot. Gaz. 71:32-60.

PAECH, K. 1955. Colour development in flowers. Ann. Rev. Pl. Phys. 6:273-298.

ROBINSON, M. G. and R. ROBINSON. 1933. A survey of anthocyanins III. Notes on the distribution of leuco-anthocyanins. Biochem, J. 27:206-212.

———— 1934. A survey of anthocyanins IV. Biochem. J. 28:1712-1720.

ROSENHEIM, O. 1920. XXI Observations on anthocyanins. 1. The anthocyanins of the young leaves of the grape vine. Biochem. J. 14:178-188.

SIMMONDS, N. W. 1954. Anthocyanins in bananas. Nature. 173:402-403.

STONE, E. C. and J. W. DUFFIELD. 1950. Hybrids of sugar pine by embryo culture. J. For. 48:200-201.

WRAY, J. 1670. Extract of a letter written to the publisher. Phil. Trans. Roy. Soc., London. 5:2063-2066.

Taxonomy of the Cenangium Dieback Fungus

BY
HUGO A. FERCHAU
T. W. JOHNSON, JR.

FINK (1911), and Weir (1921) suggested that Cenangium dieback of pines was rarely a parasitic disease, but Baxter (1937) reported that the disease was serious on exotics. Negative results from a series of inoculations of *Cenangium ferruginosum* on *Pinus sylvestris*, *P. nigra corsicana*, and *P. nigra austriaca* led Van Vloten and Gremmen (1953) to conclude that the fungus was entirely saprophytic. It has been generally stated that the severity of the disease is concomitant with a reduction in host vigor, but no studies directed specifically toward the determination of the factors responsible have been made. The fungus, *Cenangium ferruginosum*, is relatively common in the Duke Forest, but there are no available estimates of damage, nor are there any indications that the fungus is parasitic even on pines on poor sites. This fact suggested that a study of the pathogenicity of the fungus be made. In attempting to identify our collections of *Cenangium* from *Pinus resinosa* and *P. echinata*, considerable difficulty was involved, particularly in separating *C. ferruginosum* (*C. abietis*) from *C. atropurpureum* (Cash and Davidson, 1940). This paper is a report on the taxonomic aspect of a larger problem to determine the factors involved in Cenangium dieback.

There has been some confusion in nomenclature with regard to the dieback organism. The fungus was first described by Fries in 1818 and later (1823) redescribed in Systema Mycologicum as *Cenangium ferruginosum*. *Peziza abietis* is pre-Friesian, hence this name as well as combinations of it cannot be used without conflicting with the Rules of the 7th Botanical Congress (Stockholm, 1950). *Cenangium abietis* (Pers.) Duby and *C. abietis* (Pers.) Rehm, therefore, must be rejected since *Peziza abietis* is the basonym. The correct citation is *C. ferruginosum* Fr. ex Fr. Exsiccati of the collection by Rehm was examined, and although this material is labeled *C. ferruginosum*, it unquestionably represents a portion of the collection which led Rehm to change Persoon's *P. abietis* to *Cenangium*. We are following, therefore, Rehm's interpretation of the Friesian type (Rehm, 1896) and utilizing his collection as the basis for our discussion of the similarity of *C. ferruginosum* and *C. atropurpureum*. Both Rehm's collection and his interpretation of *C. ferruginosum* agree precisely with Fries' description. The taxonomic concept of *Cenangium*, on the generic level, is admittedly confused (Nannfeldt, 1932; Van Vloten and Gremmen, 1953), but need not be discussed here since the problem re-

Department of Botany, Duke University, Durham, North Carolina. We are indebted to Drs. D. P. Rogers, I. Mackenzie Lamb, C. G. Shaw, and J. S. Boyce for their cooperation in obtaining exsiccati. Special thanks are due Dr. H. Van Vloten for suggestions and comments.

ported on in this paper is at the species level. The status of *Cenangium* and related genera such as *Crumenula, Encoelia, Godronia* and *Scleroderris* is treated thoroughly by Van Volten and Gremen. We agree with their conclusions regarding the acceptability of *Cenangium* as a valid genus.

Somewhat over 160 exsiccati were examined in addition to the collections of the dieback fungus in the Duke Forest. The exsiccati represented collections throughout the United States, and several localities in Europe and Japan. Type material of *Cenangium atropurpureum* was available for direct comparison with the exsiccati. All fruiting material was treated with 2 percent KOH to swell the asci and ascospores and to extract any KOH-soluble pigment. The latter feature of *C. ferruginosum* is apparently an important diagnostic characteristic. Fresh and dried KOH-treated apothecia of one collection of the dieback fungus was examined to determine whether or not the KOH induced any variations in ascus-ascospore size. No variations were observed. Each collection was examined with regard to apothecial size and arrangement (that is, clustered or scattered), apothecial pigmentation, and ascus and ascospore size.

Cash and Davidson (1940) pointed out that *Cenangium ferruginosum* (*C. abietis*) could be distinguished from *C. atropurpureum* by apothecial size and excipulum coloration. Presumably, these investigators compared *C. atropurpureum* with exsiccati

TABLE 1. Variations in apothecial size, pigmentation, and ascus-ascospore size in some collections referable to Cenangium ferruginosum or C. atropurpureum.

Host	Apothecial size (mm.)	Pigmentation	Ascus size (μ)	Ascospore size (μ)
P. sylvestris	1-2	Purple-red[1]	68-75 × 8-11	9-11 × 5-7
P. nigra	2-3	Purple-red[2]	70-85 × 9-12	9-11 × 5-8
Abies sp.	¼-½	None	.....	.....
Picea engelmanni	½-1½	Tan	80-94 × 7-10	11-13 × 5-7
Picea engelmanni	½-¾	None	.....	.....
Picea engelmanni	1-2	Tan	110-119 × 8-10	8-9 × 6-7
Pinus sp.	¾-2½	None	77-93 × 9-12	7-9 × 4-7
Pinus sp.	¾-1	Red-brown	95-112 × 10-12	8-10 × 6-8
Pinus sp.	1-1½	None	59-70 × 5-7	11-15 × 5-7
Pinus sp.	½-1	Dark red	85-96 × 10-12	.....
Pinus sp.	¾-2½	Red-brown	70-82 × 10-14	10-12 × 5-7
Pinus sp.	1-1½	Dark red	87-98 × 11-15	.....
P. banksiana	½-¾	Purple-red	71-86 × 9-11	9-11 × 5-8
P. banksiana	1½-2½	Purple-red	82-94 × 10-14	10-12 × 5-7
P. cembroides	¾-1	None	80-90 × 8-10	9-11 × 4-6
P. contorta	1-2	Red-brown	96-112 × 8-11	.....
P. contorta	2-3	None	96-110 × 9-12	7-10 × 5-7
P. contorta	2-3½	Olive-green	121-136 × 14-17	14-16 × 6-8
P. densiflora	1-1½	Red-brown	71-85 × 10-14	11-15 × 7-9
P. densiflora	1-1½	Dark red	96-112 × 10-14	8-11 × 5-7
P. echinata	1-2	Purple-red	85-108 × 11-14	10-12 × 5-7
P. jeffreyi	1-1½	Purple-red	65-76 × 8-12	8-12 × 4-7
P. jeffreyi	¾-1	None	88-110 × 5-7	10-15 × 4-6
P. jeffreyi	1-1½	None	98-125 × 9-12	10-15 × 6-8
P. jeffreyi	¾-1¼	Purple-red	78-88 × 11-14	11-14 × 8-10
P. jeffreyi	1-2	Yellow	86-98 × 12-14	9-11 × 5-7
P. monticola	1½-2	Olive-green	.....	14-16 × 6-8
P. monticola	¾-1½	Olive-green	113-128 × 11-16	10-16 × 6-8
P. mugo	¾-1	Dark red	52-71 × 9-11	7-10 × 4-7
P. mugo	1-1½	Orange-red	91-118 × 11-15	10-12 × 5-7
P. nigra	1-2	Orange-red	62-78 × 9-12	8-11 × 6-9
P. nigra	¾-1	Purple red	104-112 × 8-11	8-10 × 6-8
P. palustris	¾-1	None	75-88 × 9-12	9-14 × 6-8

of *C. ferruginosum*, although this is not stated in their publication. One of the more recent diagnoses for *C. ferruginosum* (see *C. abietis*, in Seaver, 1951: 298) includes red-brown apothecia which are 1-1.5 mm. in diameter; the purplish-brown apothecia of *C. atropurpureum* are 2-5 mm. in diameter. Since the principal differences are those of apothecial size and coloration, these features were stressed in our observations. All specimens were compared with Rehm's *C. abietis* (the name he applied to Fries' type, *C. ferruginosum*), and type material of *C. atropurpureum*. The data in Table 1 are indicative of the variations found in the collections which we examined.

Apothecial size and excipulum pigmentation are in no way consistent. *Cenangium atropurpureum* clearly has a purple-red pigment, and, according to Cash and Davidson, *C. ferruginosum* lacks this pigment although the fungus does produce rust-red apothecia. Many of the collections otherwise characteristic of *C. ferruginosum* had the purple-red pigment characteristic of the type material of *C. atropurpureum*. The range of coloration in the specimens examined, however, was rather extensive (Table 1). Some apothecia, particularly those on *Pinus strobus*, had no KOH-soluble pigment, but were otherwise mature specimens representative in other respects of either *C. atropurpureum* or *C. ferruginosum*. In two collections it was noted that apothecia

P. ponderosa	¾-1	Yellow-brown	61-74 × 11-15	11-14 × 5-7
P. ponderosa	1-1½	None	.....	.....
P. ponderosa	½-¾	Red	81-96 × 9-12	10-12 × 5-8
P. ponderosa	½-1½	Yellow-orange	70-80 × 10-14	.....
P. ponderosa	½-1½	Purple-red	71-86 × 8-10	11-13 × 5-7
P. ponderosa	½-1	Red-brown	85-96 × 10-14	9-14 × 5-8
P. ponderosa	2-3	Red-brown	52-69 × 10-14	9-13 × 8-10
P. radiata	½-¾	None	63-75 × 9-12	10-13 × 5-8
P. radiata	1-2	Purple-red	87-96 × 9-12	10-12 × 5-7
P. strobus	½-1½	None	56-68 × 9-11	7-10 × 4-6
P. strobus	½-1	None	95-110 × 12-15	9-12 × 5-8
P. strobus	1-1¾	None	70-80 × 9-12	9-12 × 5-7
P. strobus	1-2½	Tan	70-80 × 9-13	9-12 × 5-7
P. strobus	1-1½	None	56-68 × 9-11	7-10 × 4-6
P. strobus	1-2	Yellow-brown	88-96 × 11-15	.....
P. strobus	2½-3½	None	55-65 × 9-11	7-11 × 4-7
P. sylvestris	1-1½	Purple-red	67-76 × 11-14	9-12 × 5-7
P. sylvestris	1-2	Red-brown	68-75 × 8-11	9-11 × 5-7
P. sylvestris	¾-1½	Red	71-88 × 9-12	.....
P. sylvestris	1-1½	Purple-red	70-81 × 9-11	9-11 × 5-7
P. sylvestris	2-3	Purple-red	98-120 × 14-16	9-12 × 5-7
P. sylvestris	1-1½	Red-orange	64-83 × 9-12	9-13 × 5-8
P. sylvestris	1-2	Red	68-75 × 8-11	9-11 × 5-7
P. sylvestris	1-1½	Purple-red	56-64 × 9-11	10-12 × 7-9
P. tabulaeformis	1½-2½	Red-orange	.....	9-12 × 6-9
P. taeda	2-4	None	98-112 × 11-15	10-15 × 6-9
P. taeda	2-3	Red	75-85 × 10-12	10-12 × 5-8
P. taeda	1-2	None	55-70 × 9-11	6-8 × 4-6
P. taeda	1½-2	Purple-red	92-110 × 11-15	10-14 × 5-8
P. taeda	1-2	Red-brown	78-86 × 10-12	12-16 × 8-11
P. taeda	1-1½	None	92-110 × 11-15	10-14 × 5-8
P. taeda	2-5	None	71-82 × 11-14	10-15 × 6-9
P. taeda	2-3	Purple-red	70-85 × 10-13	.....
P. virginiana	½-¾	Purple-red	72-88 × 10-14	7-11 × 4-6
P. virginiana	¾-1	None	56-72 × 6-9	8-12 × 5-7
P. virginiana	2-3	None	88-102 × 12-15	10-12 × 4-7
P. flexilis	1-2	Red-orange	.....	.....
Pseudotsuga menziesii	½-1	Dark red	65-76 × 10-12	8-10 × 5-7
Pseudotsuga menziesii	½-1	None	.....	9-11 × 7-9

[1]Identified as *C. ferruginosum* Rehm.
[2]Type material, *C. atropurpureum* Cash and Davidson.

exhibiting, macroscopically, a rust-red color had a purple-red soluble pigment; conversely, purple-black apothecia from the same collections had only a red or dark red-brown pigment.

Forty-six collections had apothecia with the purple-red pigmentation of *Cenangium atropurpureum* (and Rehm's *C. abietis*). Fourteen of the 46 had apothecial size corresponding to *C. atropurpureum*. Of these 14, however, only 2 collections (one of which was type material) had apothecia in which the asci were of a size comparable to those of *C. atropurpureum*. The remaining 12 collections produced apothecia in which the asci were intermediate in size between those reported for *C. ferruginosum* and *C. atropurpureum*. Thirty of the 46 collections with purple-red apothecia had apothecial size characteristic of *C. ferruginosum*. Seven of the 30 had asci which were of a size comparable to those of *C. ferruginosum*, 7 had asci nearly identical in size to those of *C. atropurpureum*, and 15 had asci which were considerably larger than those in the type material of Cash and Davidson's species. Judging from the literature, *C. ferruginosum* has considerably larger asci than *C. atropurpureum*. Rehm's material, however, has an average ascus size clearly comparable to that of *C. atropurpureum* (see Table 1, first two entries).

A similar comparison of the collections of apothecia with pigmentation other than purple-red gave results comparable to the foregoing ones (see Table 1). We were unable to detect any correlation between apothecial size, pigmentation, and ascus size.

In *Cenangium ferruginosum*, the ascocarps are reported to be clustered, gregarious, or heaped, whereas in *C. atropurpureum* the fructifications are scattered or in groups of 2 and 3. The majority of collections examined by us had apothecia which were both scattered and clustered, with only a very few collections showing either scattered or clustered ascocarps. In several collections, apothecia from small twigs were clearly gregarious, whereas apothecia on the same host but from bark of the main trunk were scattered or at most in groups of two. We could find no correlation between apothecial arrangement and any other key characteristic of either *C. ferruginosum* or *C. atropurpureum*.

Further evidence for the similarity of *Cenangium atropurpureum* and *C. ferruginosum* is found in host range. Hosts common to both species are: *Pinus taeda*, *P. nigra*, *P. virginana*, *P. sylvestris*, *P. mugo*, and *P. echinata*. *Cenangium atropurpureum* has also been reported on *P. pungens*, *P. caribaea* (*P. elliottii elliottii?*), and *P. rigida*, but these hosts have not been reported for *C. ferruginosum*. Comparing collection data of the exsiccati with the host range of *C. ferruginosum* according to Weiss (1953) shows that the range of this fungus should be extended to include the following hosts: *P. banksiana*, *P. ponderosa arizonica*, *P. mugo mughus*, *P. mugo rostrata*, *P. contorta*, *P. palustris*, *P. tabulaeformis*, *P. radiata*, *P. cembroides*, *P. elliottii*, *Picea engelmanni*, and *Pseudotsuga menziesii*.

The data in Table 1 need no further clarification. In our judgment, *Cenangium atropurpureum* is merely a variant of *C. ferruginosum*, the latter name having priority. *Cenangium ferruginosum* is a markedly variable species with the variations in ascocarps most likely being expressed as a partial function of host (substratum), maturity of the fructification (pigmentation), and certainly as a result of environmental conditions at the time of fruiting. *Cenangium atropurpureum* is not a valid species and is synonymous with *C. ferruginosum* Fr. ex Fr.

Literature Cited

Baxter, D. V. 1937. Development and succession of forest fungi and diseases in forest plantations. Univ. Mich. School For. Cons. Circ. 45 pp.

Cash, Edith K., and R. W. Davidson. 1940. Some new species of Ascomycetes on coniferous hosts. Mycologia 32: 728-735.

FINK, B. 1911. Injury to *Pinus strobus* caused by *Cenangium Abietis*. Phytopath. 1: 180-183.

NANNFELDT, J. A. 1932. Studien ueber die Morphologie und Systematik der Nicht-lichenisierten Inoperculaten Discomyceten. Nova Acta Regiae Soc. Sci. Upsaliensis, Ser. IV 8(2): 1-368.

REHM, H. 1896. Ascomyceten: Hysteriaceen und Discomyceten. *In* Rabenhorst., Kryptog.-fl. Deutschland, Oesterreich und der Schweiz 1(3): 1-1275.

SEAVER, F. J. 1951. The North American Cup-Fungi (Inoperculates). Publ. by the author, New York.

VAN VLOTEN, H., and J. GREMMEN. 1953. Studies in the Discomycete genera *Crumenula* de Not. and *Cenangium* Fr. Acta Bot. Neerlandica 2: 226-241.

WEIR, J. R. 1921. Note on *Cenangium Abietis* (Pers.) Rehm on *Pinus ponderosa* Laws. Phytopath. 11: 166-169.

WEISS, F. 1953. Index of plant diseases in the United States. Plant Dis. Surv. Spec. Publ. 1(5): 809-1192.

Argentine Woods and Forests

Maderas y Bosques Argentinos. By Lucas A. Tortorelli. Buenos Aires, Editorial Acme, S.A.C.I., Maipu 92, 1956. xxvii + 910 pp. $350. m/arg.

Review by Tom Gill
Charles Lathrop Pack Forestry Foundation

The publication of "Argentine Woods and Forests" (Maderas y Bosques Argentinos) by Lucas A. Tortorelli marks a major step forward in Latin American forestry, and places Argentina in the vanguard of Latin American Republics with respect to a systematic knowledge of her forest types and the uses of her forest products. The fruit of fifteen years of labor, it is by far the most exhaustive and informative book having to do with Latin American forest distribution and wood technology. It is also an excellent job of bookmaking, containing over 900 pages, 104 illustrations, 111 plates of microphotographs, and a bibliography of over 350 titles.

Its author, Lucas A. Tortorelli, is an internationally recognized forest authority. He holds the title of Agronomy Engineer from the University of Buenos Aires, received in 1933. He has lectured in Brazil and other countries, and was awarded a fellowship to complete his forestry training by study in France, Italy, and Finland. He has been Argentine delegate to many international forestry congresses, and was chairman of the section on silviculture at the Third World

Forestry Congress held in Helsinki in 1949. He was leader of the Argentine delegation at conferences of experts on paper and pulp, and upon the creation of the Argentine Forest Service in November 1943 served as its first director, and later became Director-General of Forests. Tortorelli was largely responsible for the creation of the law of 1948 for the protection of the forest resource, and was in charge of the national parks of Argentina, and president of the FAO Latin-American Forestry Commission.

"Maderas y Bosques Argentinos" amplifies an earlier work of Tortorelli's, published in 1940 under the title "Maderas Argentinas," which was essentially a descriptive study of the principal species of the country.

The book is divided into six chapters. Chapter I, Argentine Forests, is largely descriptive of the various timber types, their location, distribution, and composition. The second and third chapters are devoted to a technological study of the principal species, for of the 1500 woody species found in Argentina, Tortorelli has confined his study to those most valuable to commerce and industry. Under each family he discusses the geographic distribution and botanical characters of the various species, their cultural demands, the macroscopic and microscopic aspects of the woods, together with the mechanical characteristics and uses. Of especial value here is his discussion of the present uses of wood and their possible future use as substitutes for foreign species.

Chapter IV is given over to a key to tree identification based on the wood characteristics. Chapter V is made up wholly of microphotographs with brief descriptions of the species discussed. Chapter VI is composed of a bibliography and indices of illustrations and common and scientific names.

Those of us who have worked in Central and South America realize the immense need for dependable methods of identification before we can hope for an end to the chaotic conditions found today throughout the wood industries of Latin America. Confronted with the great complexity of tree species, where the same common name may be shared by a dozen different species, and where the same species in different localities may have as many different names, the stage is set for confusion, irresponsible sales, and the frustrations that attend much buying and selling of timber in those regions. Inevitably many species are sold for purposes for which they are not adapted, and no purchaser can be certain he is getting what he pays for, with the result that buyers, in despair, turn from their local species to imports. Tortorelli's book should go far in remedying this condition. It should be of immense practical value to the wood users of the Argentine and to countries importing wood from that country.

Tortorelli has pointed out that World War II had favorable consequences for the wood user of the Argentine, because it caused the forest products industry to seek out new uses for their indigenous species when they were restricted by war from importation. Bitter economic necessity caused them to devise uses better adjusted to the characteristics of many species which until a few years ago were unknown. In not a few cases it was found that Argentine woods formerly ignored by industry, or utilized for unimportant local purposes, served better than imported woods. This has had a salutary effect upon the wood industry of the Argentine, and should mark a large step forward toward freeing the industry from its past dependence on importations. From a forestry standpoint it is equally important, since by utilizing a greater number of species, forest management is made practical.

"Maderas y Bosques Argentinos" will be of dual service to the Argentine: it should help clear up what Tortorelli calls a "veritable anarchy" — a chaotic condition of confusing names that lends itself to fraud and misunderstanding—and it should bring greatly needed light regarding the uses of many indigenous woods that could well take the place of imported species.

It is an impressive, scholarly, and intensely practical contribution to professional forestry and to the forest products industry.

Sampling Methods and Community Attributes in Forest Ecology

BY
ALTON A. LINDSEY

WHEN TWO GROUPS are working with the same material, even though from different viewpoints, it is helpful to each to keep posted on what the other is doing. This applies between the fields of silviculture in forestry and plant ecology in botany. Substantial progress has been made recently in both fields toward improved methods of forest sampling. In studying the same forest stand, an ecologist would have in mind the question "what is there and why so?" while the forester would prefer answering "what is there and so what?" The second half of each question determines the manner of answering the first half, so that the resulting descriptions differ. Also, the ecologist is much more concerned with the "lesser vegetation" than is the forester. Although the objectives of ecology and forestry differ, both methodology and research results overlap to a considerable degree, and there is exchange of ideas in both directions. It may, therefore, be of interest to those foresters who have not followed the voluminous literature of plant sociology to have a brief review of recent methods and concepts applicable to the tree stratum. It should be stressed that this paper deals with ecological literature, rather than forestry literature.

In vegetational studies as elsewhere, analysis separates the elements of a complex and in the process brings into sharper focus the relationships between them. Ecology deals not only with the "morphology" of the organized plant community, but also with the community-level "physiology" of its operating processes. Ecological meaning lies chiefly in the relationships between structure, function, and history.

Present interest in methods and the statistical approach, in the statistician's sense, is strongest in the English-speaking countries. Empirical work by ecologists on artificial and natural populations has been supplemented by theoretical studies by mathematicians and ecologists, in a search for relationships and in developing refined methods for the more abstruse attributes such as the nature of the dispersal of a species in the stand. The most frequent use of statistics is to determine adequacy of sampling. On the other hand, workers who stress the characteristic or faithful (though perhaps infrequent) species rather than the abundant ones, rely on the species-area curve rather than statistical methods for determining sampling adequacy.

What is the place of quantitative methods in vegetation study? Measuring things can be used as an aid in understanding them, or as a substitute for understanding them. Properly used, quantitative methods may contribute much in the long run to the classification of vegetation. More im-

---

The author is associate professor of plant ecology in the Department of Biological Sciences, Purdue University, Lafayette, Indiana.

portant than classification, in the view of American ecologists at least, is the solution of environmental problems. The sampling of communities furnishes reasonably objective data of value in a variety of problems in botany and applied work in forestry, wildlife management, range management and soil conservation. Several other fields farther removed from orthodox ecology, such as engineering and military applications and land-use planning, may be satisfied with broader, less precise results of vegetational techniques.

Selecting stands for sampling, and placing limits on the allowable intrastand diversity to be lumped together in the data are much more important and difficult matters in ecology than in forest inventory. The interpretive judgment of the worker takes precedence over any method that can be applied before sampling is started. This is true even when some objective randomization technique is used for selecting stands; there are almost always completely different types within the general bounds of the region studied, and these places cannot be considered as grist for the randomization. A broad stand of a particular rank may be further broken down into a very restricted rank, depending on the purpose and viewpoint. As such finer sampling proceeds, where to draw the line remains a matter for repeated subjective decisions. This source of subjectivity is minimized when working with the broader hierarchical groups or a more general interpretation of what may be included in a single community. As long as people have different opinions, vegetational sampling will be partly art, not wholly science.

To be studied or sampled as a representative of a single community type, any vegetational unit is required to possess a degree of homogeneity. An attempt may be made to decide this from observation. However, statistical tests for homogeneity have been and are being developed. Data collected from the stand are subjected to such a test; if the stand falls below a certain standard on the scale of homogeneity the data are not utilized further.

Among the considerations that influence the choice of a sampling method is the allowable scope of variation to be included in one set of data. When minor differences within small areas are to be discriminated but only one or relatively few of the recognizable subdivisions are being studied, small quadrats or the point methods using distance measures are most suitable. Of course these methods are not restricted to such fine breakdown. The line-interception and line-strip methods as commonly used lump more of the variation, integrating each diverse condition, in its proper proportion, into a picture of a stand entity in a broader sense. However, by using a linear unit as a transect is used in studying zonation (instead of as a single quadrat is used) and recording the recognizable vegetational boundaries on the line, linear methods are quite suitable for detailed mosaics where most or all types making up the pattern are being sampled for study. When so used, linear methods have the pronounced advantage over point or quadrat methods that they measure rather accurately the ground areas occupied by each sub-division.

Any acceptable sampling method should give information on the nature of dispersal of the several principal species. Dispersal is either random or departs from randomness toward regular, uniform spacing or toward aggregation. Clark and Evans (1954) stated "In a random distribution of a set of points on a given area, it is assumed that any point has had the same chance of occurring on any sub-area as any other point, that any sub-area of specified size has had the same chance of receiving a point as any other sub-area of that size, and that the placement of each point has not been influenced by that of any other point." They stressed that randomness is a spatial concept, intimately dependent upon the boundaries of the space chosen by the investigator.

TABLE 1. Terms for the definition of the attributes in Table 2. Symbols referring to "areal plots" apply to quadrats and the strip component of line-strips; those referring to "lines" apply to line interception used alone or to the line component of line-strips. Two point methods are included — the point-centered quarter (QR) and the Bitterlich method (VR). Each stand is treated separately.

N	no. of individuals of a species in all areal plots (or at all points)
N_t	total no. of individuals of all species in all areal plots (or at all points for D_3, B_8, and B_9)
N_p	no. of individuals of a species at all sampling points (VR)
N_{pc}	no. of individuals of a species in a given size class at all points (VR)
n	no. of areal plots
n_s	no. of areal plots in which a given species occurs
n′	no. of lines used
n_p	no. of points (VR)
a	area of one of the areal plots in square feet
F_t	total of frequencies for all species
I	sum of interception distances for a given species on all lines
I_t	total interception distance for all species on all lines
L_t	total length of all lines collectively
d	median diameter (in inches) of the size class being considered (VR)
b	sum of basal areas for a species in all areal plots
b_t	total basal area for all species in all areal plots
m	mean distance (over all points used) between a point and the trunk center of the nearest tree in its quarter irrespective of species (QR)
A	10 divided by the basal area of a trunk having diameter d (which see) (VR)

Quantitative Phytosociological Attributes

In order to compare different methods, the vegetational characteristics to be studied thereby should first be considered. Table 1 defines the terms which are used in the formulae for the various parameters in Table 2. Each parameter is symbolized by a capital letter, indicating its vertical column, followed by a subscript number representing the horizontal row. This set of terms and symbols and the formulae utilizing them complete the system which was introduced only partially in a paper (Lindsey, 1955) restricted to one sampling method. Relationships between the concepts are brought out by the tabular arrangement. Only four sampling methods (quadrat, quarter, angle-count or variable plot-radius, and line-interception) were considered, but combinations of these were covered thereby. Attributes for which more than one stand is required are omitted. The concepts are generally familiar to ecologists and in current use by them, except for the two new expressions in column "O". Some are merely means for obtaining others. Data for expressions throughout the subscript 1 row are not comparable unless obtained from plots of the same area; elsewhere in the table, values for any given plot-derived attribute are comparable regardless of plot size.

The fundamental density concept, density per small unit area (D) is distinguished from the more practical D_2 or density per large unit area. Feet and acres are preferred in the United States over meters and hectares. Because the two least important density concepts have been omitted to simplify the table, subscript 6 and 7 rows are not shown. The existence of nine distinct concepts of density shows that the use of this term without definition is worse than ambiguous.

Below the double line the attributes (8, 9, and 10) do not call for discrimination of species. These attributes are of more use in forestry, engineering, and military applications than in straight ecology, and can often be studied from airphotos. Unfortunately, species identification of individual trees is possible from airphotos only in certain vegetation types or under special

TABLE 2. Terminology, definitions, and relationships among intrastand attributes for forest ecology. (For terms used see Table 1.) Parameters above the double line refer to a given species separately, those below it refer to the tree population as a whole irrespective of species.

Sub-script	D	M	A	B
	density per sq. ft. $D = N/na$	mean area (sq. ft.) per indiv. $M = na/N = 1/D$		
1	plot density $D_1 = N/n$		plot abundance $A_1 = N/n_a$ $= 100\ D_1/F_1$	basal area (sq. ft.) per plot $B_1 = b/n$
2	density per acre $D_2 = 43560\ D$ $=$ sum of D_5 [VR] $= D_3(D_0)/100$ [QR]			basal area (sq. ft.) per acre $B_2 = 43560\ b/na$ $= 10\ N_p/n_p$ [VR] $=$ sum of B_5 " $= B_3(B_0)/100$ [QR]
3	relative density $D_3 = 100\ N/N_t$			relative basal area $B_3 = 100\ b/b_t$
4				
5	density per acre for a given size class of 1 species $D_5 = \frac{B_5}{.00545d^2}$ [VR] $= (N_{pc})k/n_p$ "			basal area (sq. ft.) per acre for a given size class of 1 species $B_5 = 10\ N_{pc}\ /\ n_p$
8	stand density per sq. ft. (see D_0 and M_8) $D_8 = N_t/na$	stand mean area (sq. ft.) per indiv. $M_8 = m^2$ [QR] $= 1/D_8$		stand basal area (sq. ft.) per indiv. $B_8 = b_t/N_t$ [QR]
9	stand density per acre $D_0 = 43560\ N_t/na$ $=$ total D_2 for all species $= 43560/m^2$ [QR]			stand basal area (sq. ft.) per acre $B_0 = 10\ N_t$ [VR] $= B_8(D_0)$ [QR] $=$ total B_2 for all species
10				

conditions. This fact drastically limits the usefulness of airphotos in plant ecology.

Columns D and A indicate numerical abundance, F is determined by both numerical abundance and the nature of dispersal, while M, B, and C reflect areal relations.

The only formulae that call for special mention are those for "prevalence" and "predominance" in the final (O) column.

TABLE 2. Continued.

Sub-script	C	F	Y	O
1	line cover $C_1 = I/n'$	frequency $F_1 = 100\ n_s/n$		
2	cover (sq. ft.) per acre $C_2 = (43{,}560)\ C_4$			
3	relative vegetation cover $C_3 = 100\ I/I_t$	relative frequency $F_3 = 100F_1/F_t$	Importance $Y_3 = \frac{B_3+D_3+F_3}{3}$	Prevalence $O_3 = \frac{(B_3+C_3)(D_3+F_3)}{400}$
4	percentage ground cover $C_4 = 100\ I/L_t$			Predominance $O_4 = \frac{(B_3+C_3)(D_3+C_4)}{400}$
5				
8				
9	stand ground cover per acre $C_9 =$ total C_2 for all species $= 43{,}560(C_{10})$			
10	stand percentage ground cover $C_{10} = I_t/L_t$ = total C_4 for all species			

These are new summary expressions for integrating several parameters into one number to express general dominance or relative biotic influence of a given species in the stand. Both O_3 and O_4 require determination of crown cover, thus lend themselves best to the line-strip method. They are derived from the phytograph, a familiar graphic method of expressing the community status of a given species. A phytograph is drawn within a circle having radius 100, by plotting percent values for four attributes on the four main radii, then connecting the points by four straight lines. The O_3 and O_4 values are based on the areas within these four-sided phytograph polygons of two types, O_3 using the frequency percentage as one of the four, O_4

substituting ground cover (C_4) on that radius. Prevalence (O_3), then, is a per cent value showing the area of the phytograph figure (based on four subscript 3 parameters) relative to the area the polygon would have if each component had a radius of 100. Using the terms from Table 2, the prevalence concept is derived thus:

$$O_3 = \frac{\frac{B_3(D_3+F_3)}{2} + \frac{C_3(D_3+F_3)}{2}}{200}$$

and is computed by the equivalent simpler formula given in space O_3 in Table 2. A species forming a pure stand, and occurring in all areal plots used, has the maximum O_3 value of 100.

Because of its double weighting of cover by use of both vegetation cover and ground cover, predominance (O_4) appears to be the best integrative index. Its formula is shown in space O_4 of Table 2. The theoretical maximum value (100) would obtain if the single species of a pure stand were to cover all the area of ground within the plots used, or to intercept the total length of all lines.

Sampling Methods

The familiar quadrat has been used for many years in forestry and ecology. It is an areal sample unit of square, rectangular or circular shape, and of a size suitable to the community or stratum being sampled. Curtis and McIntosh (1950) discussed quadrat methodology, reviewing the attributes obtainable by quadrating and the interrelationships among them. Their concepts and terminology are followed herein for the most part.

The quadrat method yields all of the parameters from within a single stand except cover (column C), which it is impracticable to obtain from quadrats, especially in forests. In contrast, the line-intercept method, applied first to grassland and shrub communities, and only recently to trees, estimates cover expressions reliably but is seriously biased for any other attributes. Although crown cover is the most important single parameter of a species in its community relations, it is not measured directly by most ecologists; instead tree basal area (B) at 4.5 feet from the ground is taken. The basal area idea was borrowed from forestry, where it is of obvious value for bole volume estimates; it is widely used in forest ecology because basal area roughly parallels crown cover. The latter is the major determinant of "dominance" or the influence which the species exerts in coaction and reaction. Since the assumed parallelism sometimes fails, crown cover would be measured in preference to basal area were it not usually relatively difficult to obtain in forests of tall trees. The line-strip method (Woodin and Lindsey, 1954; Lindsey, 1955) was designed to facilitate estimating all important parameters including cover. The line-strip sampling unit is a combination of a long, narrow areal plot with line interception, but only the line is surveyed. The line yields cover values by downward projection of crown interception points to a steel tape by the use of a level-bubble on a sighting bar. The strip or elongated quadrat yields all other attributes, of which frequency is computed from fractional parts of the strip. The operator works the length of the 400 or 600 foot line-strip once to obtain all measurements. A set of 6 to 8 such sampling units are needed at each station, ordinarily, for adequacy. The method is considerably slower than the rapid survey methods discussed below, but includes the important coverage parameter.

In contrast to the relatively slow techniques centered around the quadrat method, stands a group of spacing or distance methods developed chiefly by Cottam and Curtis which are based on randomly located points. The first of these was the random pairs method, in which the one tree nearest the point, and another nearest the point on the other side (outside a specified exclusion angle), are measured for basal area and their distances to the point recorded. Much research has been devoted to testing the

various distance methods, and it appears that the best of these is the point-centered quadrant or quarter (Cottam *et al.*, 1953). At a random point in the stand, an imaginary cross is centered, and the distance from the point to the nearest tree in each quadrant is measured by tape and its basal area also determined. Formulae marked "QR" in Table 2 show how density is obtained in this method. A modification by Cottam is to measure the distance from point to tree with a land range-finder having a six-inch optical base.

Bitterlich (1947, 1948), an Austrian forester, introduced an ingenious point method termed the "variable-plot radius method," or "angle-count method." The latter name seems confusing to the ecologist because of the various other angle methods (Morosita, 1954) related to the random-pairs and quarter methods. Bitterlich's method is plotless in practice but not in theory. Since Grosenbaugh (1952) published on the method, it has been well known, so that it need not be described herein.

Since Bitterlich's method is excellent for obtaining basal area estimates, it is regrettable that basal area does not have as high a value for phytosociology as it has for forestry. However, it is a needed ecological attribute, especially when the more valuable one, crown cover, is disregarded, as in this method. The possibility of full operation from the point is advantageous except when specimens must be examined closely to ascertain species identification.

Density determination by this method is rather roundabout. The second formula given under D_5 in Table 2 is the more convenient one; the resulting figures for different size classes are added to obtain species density. Rice and Penfound (1954) considered the variable-plot radius method insufficiently accurate for density, and have applied a combination of this with the quadrat method, which they term the augmented variable-radius method. This speeds quadrating by use of paced arm-length-wide strips instead of more accurately surveyed plots, and limits Bitterlich's method to obtaining basal area, for which it excels.

Toynbee (1946) used the term "etherealization" to characterize the evolution of techniques in general, in that refinements tend to bring about reduced dependence on material factors. In communications, for example, radio and television are more ethereal media than the older telegraph or telephone. Etherealization in phytosociology is evident in the attempt to mitigate the difficulties of quadrating or to bypass the quadrat method altogether. One spur to simplifying sampling procedure has been disillusionment concerning some of the traditional accompaniments of the quadrat method. Various workers have lost faith in the value of the species-area curve and in the universal applicability of the 10×10 meter quadrat size for forest trees. Statistical work has brought out the large aggregate number for each species that must be included to give reliable results, which adds to the burden of quadrating. A consideration by Cottam *et al.* (1953) of the characteristics of random populations shows the necessity of thorough sampling even with randomness, and this is all the more true for aggregated species.

Most of the sampling methods fall into two broad classes, detailed methods *versus* rapid-survey methods. Detailed methods include the quadrat and the line-strip method. The augmented variable-radius method is an effective compromise between detailed and rapid methods. Rapid survey techniques include various spacing methods, line-interception, and the variable plot-radius method.

These methods form a series of increasingly ethereal and simple techniques, requiring progressively less material equipment or time-consuming effort. This is illustrated in a statement by Rice and Penfound (1955): "In deciding on a method, three considerations were paramount: (1) the method must be rapid, (2) it should require little equipment, and (3) it must include data for frequency, density, and

basal area." The quadrat as usually applied falls down for the first two of these requirements, while more ethereal methods meet them. Shanks (1954) adds two additional criteria: "operable by one person at a reasonably efficient level, and susceptible to statistical measures of variability so that confidence limits can be established."

The Bitterlich method is the most ethereal one that has been used by plant ecologists in published work to date. Grosenbaugh suggested the use of a low power monocular calibrated to show the angle. Another etherealization is Bruce's (1955) suggestion, whereby the only equipment needed is a wedge prism of 3 diopters. In our experience in Indiana hardwoods, the original angle gage bar, although more cumbersome to carry, is easier and more accurate in sighting than the prism.

Now that a high degree of etherealization has been reached, are we justified in abandoning the detailed methods that utilize sampling units of known area? This is dubious, because the rapid methods have certain inherent disadvantages. (1) As a general rule, the more ethereal a rapid method is, the fewer the vegetational parameters obtainable. On the other hand, etherealizing the quadrat produced another detailed method, the line-strip, that is faster than quadratting, and yields everything the quadrat does and coverage besides. (2) Most of the rapid methods are well suited only for the tree stratum, particularly non-reproduction class trees. There is a strong temptation to disregard all other plants in the community. (3) Increasing simplification of field procedure may be accompanied by increasing complexity of the underlying theory, as in Bitterlich's method. (4) A related disadvantage involves statistical confirmation of sampling adequacy, because there had been little mathematical study of the rapid methods until recently, and a probability theory for them had not been worked out. Consequently, in using these methods to date, sampling adequacy has seldom been demonstrated statistically. This has now been potentially overcome by mathematical study for most of the rapid methods. The line-intercept coverage method is included by J. A. Navarro in his unpublished Ph.D. thesis in mathematics (Purdue Univ., 1955). Morosita (1954) has published on the theory of spacing methods in Japan, but in English. As a result we know that a given statistical expression, such as percent standard error, is essentially comparable among these methods and the quadrat method. However, much mathematical study is needed on the probability theory involved in the variable plot-radius method. Until this is available, it is probably unjustifiable to compare statistical expressions for density results with corresponding ones derived from other sampling methods.

Carrying etherealization to its logical extreme would bring us full circle, back to the simplest and earliest method, the naked estimate or educated guess. This requires no equipment, the barest minimum of time in the field and in computation. In reality, it is no mere *extensio ad absurdum*. Estimation is of course much used in range ecology. It puts high demands on the operator's experience. Although few would consider that walking casually through a stand is a proper phytosociological method, an ecology graduate student who had done forestry work in the type, thus made density estimates that checked well with a complete tally in a red pine—white pine—jack pine stand. The average of his percent errors for the 3 dominant species was 5 percent. Compare this with a 13% average error from quadrat sampling there, which took about 30 times as long and included 13% of the stand. If an ecologist could visually estimate with confidence the main parameters as well as that student did density in this instance, no sampling methods would be needed. Of course, lacking full census and sampling, such estimates could not be tested for confidence. The most ethereal "method" of all, requiring no equipment, is the least susceptible to validation.

In spite of lack of confidence validation, systematic routine ocular estimation deserves

wider use in descriptions of plant communities. One place for it is in recording and analysis of certain things by IBM punch cards. For rapid surveys or in supplemental descriptions, such observations as physiographic conditions, field estimates of soil texture classes, species phenology, stratum rank, and relative shading of lower strata by canopy closure, lend themselves well to coding on a 1-10 scale. Unequal scales may be used when appropriate. Such information, rapidly marked by numbers on field forms, each horizontal line to be punched later into one corresponding species card, gives much more information than a mere species list. Incidentally, recording on printed forms for later card punching is more reliable for machine analysis than recording on mark-sense cards in the field, under the conditions of most ecological work.

During rapid surveys, lower strata species may be omitted, they may be merely listed, or line interception could be used supplementally for coverage only. If one wants to avoid species identification for herbs and shrubs, useful structural data can be recorded by Dansereau's (1951) method or by the symbols of Christian and Perry (1953). As usual, methods will be selected in the light of objectives.

Etherealization, then, has given us new rapid methods which are excellent for reconnaissance and preliminary studies or extensive surveys. Another general group, revolving around the quadrat method, requires more equipment, time, and labor, but yields more complete information about the community as a whole. Hence, these less ethereal methods are still suitable for intensive, detailed work, or definitive vegetational analyses of especially interesting stands. The speed of the rapid survey methods, despite their disadvantages, is so outstanding that they are being adopted increasingly by plant ecologists.

A promising new etherealization of the quadrat method, combined with the basal area portion of the variable-radius method, yields the large number of trees per operational unit that characterizes an ample quadrat, together with over-all speed, ease of basal area determination, and density without diameter class estimation or measurement. Observations are made through the 360° at one place which serves both as a point for variable-radius tallies for basal area only, and as the central point of a one-tenth or one-fifth acre quadrat (depending on density of trees and occurrence of obscuring undergrowth). The variable-radius angle instrument is either a wedge prism, now available commercially for this purpose, or six-power monocular with an extra disc marked with short vertical lines setting off the proper angle. The monocular also facilitates species identification from the point. A six-inch base field range finder, such as Cottam recommends for measuring tree-to-point distances in quarter sampling, is fixed at the proper radius distance to accept or reject trunks for circular areal plots. This avoids surveying quadrats in the usual laborious fashion.

This combination method needs further trial but it appears that it is very rapid and avoids the weak features of the two methods while retaining their advantages. Although as well suited for one-man use as any other method, it would have its highest efficiency with one man staying at the point using the instruments, while a roving helper records data for him, checks cases of doubtful identification, and holds aside foliage that obscures the sighting. However, if the objection of Rice and Penfound to the accuracy of density by Bitterlich's method is not justified in practical work, the suggested combination method would have no advantage over using Bitterlich's method for both basal area and density.

Summary

Substantial progress has been made recently in the development of improved ecological sampling techniques for analysis of forest communities. The vegetational attributes constituting the objectives of sampling methods are redefined and their inter-

relationships indicated by means of a symbolic system presented in tabular form. The recent expansion of quantitative methods and concepts suggests the usefulness of symbols and formulae, particularly in the teaching of ecology. Two new summary expressions, prevalence and predominance, which integrate the role of cover with other parameters, are introduced.

Two general groups of methods, detailed *vs.* rapid survey methods, are distinguished. Detailed sampling techniques include the *quadrat* method involving surveyed boundaries, and the *line-strip* method —which combines a linear unit and an areal strip but in which only the line is surveyed. Rapid methods include distance or spacing methods such as the *random-pairs* and the preferable *quarter* method, and the Bitterlich *variable-plot-radius* method. The various methods are evaluated, and a new method, involving use of instruments from one point to combine variable-radius sampling with the quadrat, is suggested.

Literature Cited

BITTERLICH, W. 1947. Die Winkelzahlmessung. Allgm. Forst-u. Holzwirts. Ztg. 58: 94-96.

————. 1948. Die Winkelzahlprobe. Allgm. Forst-u. Holzwirts. Ztg. 59 (1/2): 4-5.

BRUCE, DAVID. 1955. A new way to look at trees. J. For. 53: 163-167.

CHRISTIAN, C. S. and R. A. PERRY. 1953. The systematic description of plant communities by the use of symbols. J. Ecol. 41: 100-105.

CLARK, P. J. and F. C. EVANS. 1954. Distance to nearest neighbor as a measure of spatial relationships in populations. Ecology. 35: 445-453.

COTTAM, G., J. T. CURTIS and B. W. HALE. 1953. Some sampling characteristics of a population of randomly dispersed individuals. Ecology. 31: 434-455.

CURTIS, J. T., and R. P. MCINTOSH. 1950. The interrelations of certain analytic and synthetic phytosociological characters. Ecology. 31: 434-455.

DANSEREAU, PIERRE. 1951. Description and recording of vegetation upon a structural basis. Ecology. 32: 172-229.

GROOENBAUGH, L. R. 1952. Plotless timber estimates — new, fast, easy. J. For. 50: 32-37.

LINDSEY, A. A. 1955. Testing the line-strip method against full tallies in diverse forest types. Ecology. 36: 485-495.

MOROSITA, MASUAKI. 1954. Estimation of population density by spacing method. Mem. Faculty of Science, Kyushu University, Ser. E (Biology) 1 (4): 187-197.

RICE, E. L., and W. T. PENFOUND. 1955. An evaluation of the variable-radius and paired-tree methods in the blackjack-post oak forest. Ecology. 36: 315-320.

SHANKS, R. E. 1954. Plotless sampling trials in Appalachian forest types. Ecology. 35: 237-244.

TOYNBEE, A. 1946. A study of history. Oxford University Press. New York and London.

WOODIN, H. E. and A. A. LINDSEY. 1954. Juniper-pinyon east of the continental divide, as analyzed by the line-strip method. Ecology. 35: 473-489.

Tissue Differentiation and the Application of Sodium Arsenite to Girdled Trees Relative to Bark Peeling

BY
M. REINES

THE ARMSTRONG FOREST COMPANY of Johnsonburg, Pennsylvania, has developed a practical method for removing bark from standing trees. Sodium arsenite solution (Atlas "A") is applied to the peeled face of a girdled tree, when bark slippage is apparent, during the so-called "sap peeling" season. Consequently, the tree is quickly killed and the bark subsequently separates from the wood.

This investigation was undertaken to explore the nature of bark slippage during the "sap-peeling" season and the role of sodium arsenite in bark peeling when used on white oak (*Quercus alba* L.), black oak (*Q. velutina* Lam.), pignut hickory (*Carya glabra* (Mill.) Sweet, sugar maple (*Acer saccharum* Marsh), black cherry (*Prunus serotina* Ehrh.), and white ash (*Fraxinus americana* L.). Trees used in this study were located in woodlots of the Pennsylvania State University in central Pennsylvania.

Methods

To ascertain the nature of bark slippage, tissue differentiation was studied in living trees throughout the year. Specimens approximately 1½ × ½ inch and ½ inch thick, with dark, cambial zone, and wood intact were collected. To facilitate the removal of the sample some of the outer bark was removed with a draw-knife. A bark scribe was then used to cut two parallel longitudinal grooves into the wood, approximately 1 inch apart. The depth of these was increased with wood chisel and mallet. With the cutting edge of the chisel slanted away from the specimen block the top and bottom grooves were cut approximately ½ inch deep. The block thus isolated on four sides was pried out with the chisel's edge.

Collection of specimens began in June, 1951. They were taken from two trees of each species at approximately breast height from the north side and subsequently at

The author is now at the School of Forestry, University of Georgia, Athens, Georgia. He gratefully acknowledges the cooperation and financial aid rendered by the Armstrong Forest Company. Especial thanks is extended to Professors Henry W. Popp and David A. Kribs of the Department of Botany and Plant Pathology for their advice and interest throughout the progress of the investigation, and to Professor H. Norton Cope of the Forestry School and Professor Donald E. H. Frear of the Agricultural Biochemistry Department of the Pennsylvania State University for their assistance and facilities provided by them. This paper is part of a dissertation submitted to the faculty of the Pennsylvania State University in partial fulfillment of the requirements for the Doctor of Philosophy. Authorized for publication on April 20, 1954, as paper No. 1872 in the Journal series of the Penn. Agric. Expt. Sta., and as a contribution No. 186 of the Department of Botany and Plant Pathology.

TABLE 1. Approximate dates of tissue differentiation and bark slippage.

	Tree No.	D.B.H. (in.)	Height	Age (years)	Condition of buds or leaves when bark slippage begins	Initiation of xylem formation	Slippage began	Cessation of xylem formation	Total lignification of xylem	Bark slippage ceases	Initiation of phloem formation	Cessation of phloem formation
Q. alba	1	20	78	139	.....	.....	.....	8/13/51	8/27/51	8/27/51	.....	.....
	2	20	78	118	.....	.....	.....	8/20/51	8/27/51	8/27/51	.....	.....
	3	15	65	104	Buds swelling	4/11/52	4/11/52	.....	.....	.....	4/11/52	.....
Q. velutina	4	20	74	95	.....	.....	.....	8/20/51	8/27/51	8/27/51	.....	.....
	5	19	79	100	.....	.....	.....	8/20/51	8/22/51	8/27/51	.....	.....
	6	15	67	83	Buds swelling	4/4/52[1]	4/11/52	.....	.....	7/26/52	4/18/52	.....
Carya glabra	7	18	82	100	.....	.....	.....	8/13/51	8/27/51	8/27/51	.....	7/23/51
	8	20	82	100	Buds burst, young	.....	.....	8/13/51	8/27/51	8/27/51	.....	7/23/51
	9	12	75	100	leaves present	5/2/52	5/2/52	7/26/52	.....	.....	5/23/52	.....
A. saccharum	10	13	50	68	.....	.....	.....	8/13/51	8/27/51	8/27/51	.....	.....
	11	11	60	53	Bud identity lost,	.....	.....	8/13/51	8/27/51	8/27/51	.....	.....
	12	11	43	60	leaves ½ mature	5/16/52	5/16/52	.....	.....	.....	4/11/52	.....
	13	10	50	49	size	4/29/52	4/29/52	.....	.....	.....	4/11/52	.....
P. serotina	14	14	70	58	.....	.....	.....	8/27/51	9/10/51	9/10/51	.....	.....
	15	14	69	61	Bud identity lost,	.....	.....	8/27/51	9/10/51	9/10/51	.....	.....
	16	15	67	48	leaves ½ mature	5/9/52	5/9/52	.....	.....	.....	4/11/52	.....
	17	....	....	....	size	5/9/52	5/9/52	.....	.....	.....	4/11/52	.....
F. americana	....	....	....	....	.....	4/17/52	4/17/52	.....	.....	.....	5/1/52	.....

[1]South side. Date was 4/18/52 on north side.

equal intervals around the tree. Collections were made weekly until August 27, 1951, bi-weekly thereafter until December 2, 1951, and weekly after February 2, 1952, until July, 1952. During the course of sampling two additional trees of each species were used as further sources of study material. Specimens of ash were also collected, at the beginning of the "sap peeling" season.

Upon collection, specimens were immediately immersed in formalin-aceto-alcohol and subsequently washed, dehydrated, and imbedded in parlodion. Sections were cut 10 to 30 micra thick and stained with safranin and fast green to distinguish between lignified and unlignified tissue.

To observe the effects of sodium arsenite on living trees the solution was applied immediately to the peeled face of the girdle according to the practice of the Northeast Pulpwood Research Center (Anon., 1950), Ehrhart (1951), and Bennett (1951). In making the girdle the bark was removed carefully without cutting into the wood. Sodium arsenite (Atlas "A", Chipman Chemical Company) was applied to the peeled face with a paint brush. Subsequent to treatment leaves were collected for chemical analysis and samples of bark and wood were removed periodically at six-foot intervals up the trees, for histological examination and chemical analysis. Also, wood specimens from two radii at each of two levels in treated trees were analyzed chemically to determine the pattern of distribution of translocated arsenical solution. The presence and amount of arsenic were determined by the Gutzeit method.

Observations

The approximate dates of the initiation of xylem and phloem formation, the cessation of xylem formation, the completed lignification of xylem, and the beginning and ending of bark slippage are given by species in Table 1.

At the beginning of the growing season, in all species studied, the first indication of any change preliminary to the addition of

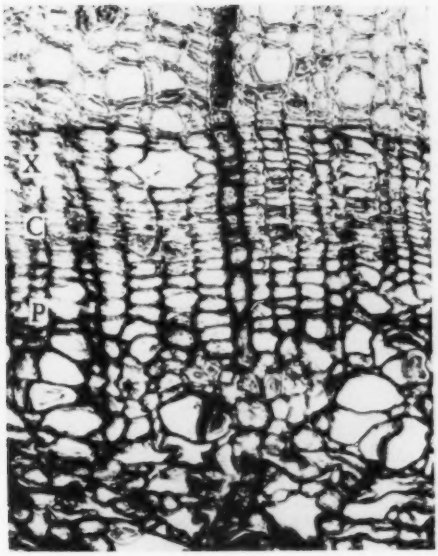


FIGURE 1. *Cross-section of white oak showing the differentiation of first young vessels of the new xylem in the new growing season; xylem (X), cambial zone (C), phloem (P).*

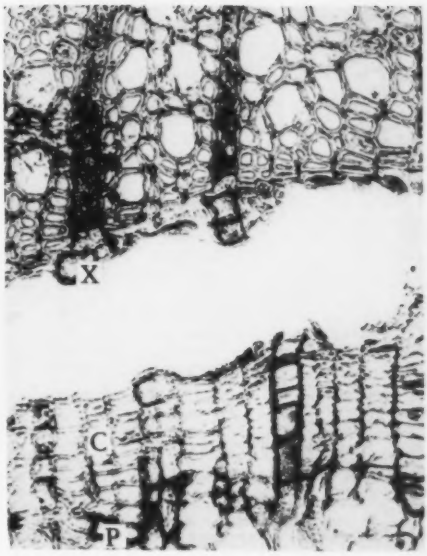


FIGURE 2. *Cross-section of white oak showing the separation of bark from wood in the region of differentiating tissues of the xylem.*

new cells by the cambial zone is the apparent swelling of its cells. They then begin to divide to form new cells of an undifferentiated tissue. Neither new xylem nor new phloem are discernible. However, soon individual centripetally located cells of the mass of undifferentiated tissue enlarge to form the first recognizable xylem cells of the new growing season (Fig. 1). This phase has been designated as the initiation of xylem formation in Table 1. Bark slippage or "sap peeling" begins at this stage and continues until maturation of the xylem is completed after the end of the growing season. During this period, when the bark is manually separated from the wood, the break occurs in the xylem where cells have enlarged and differentiated (Fig. 2). At the end of the growing season, while xylem formation stops with the cessation of cambial activity, it is not until the last-formed xylem cells undergo complete maturation, a process manifested by the secondary thickening and lignification of the cell walls, that bark slippage ceases. At this time an abrupt transition exists between the xylem and a well-defined cambial zone, which is reduced in radial thickness and number of cells.

The inception of phloem formation is marked by the appearance of newly differentiated cells located centrifugally to the cambial zone and adjacent to a tangentially oriented band of parenchyma formed during the preceding growing season. The sieve tubes of the old phloem are crushed by the outward pressure of the new in all species studied except hickory.

When sodium arsenite solution is applied it moves upward in the tree. Examples of chemical analyses made to demonstrate the movement and the presence of arsenic above and below the level of application in treated trees are listed in Tables 2 and 3. Chemical analyses of cross-sections of treated trees reveal that the greatest concentrations of arsenic lie and therefore move in the sapwood close to the bark in all species studied.

TABLE 2. Concentration of arsenic (ppm) in wood at different levels.

Test	Species	Height of tree	Treatment	Date of Treatment	Time after Treatment	Just below	Number of feet above girdle 3	6	12	18	24	30	36	42
1[1]	White oak	65	Treated side	7/12/51	10 hrs.	.....	81	44	20	0	0	0	0	
			Untreated side	7/12/51	10 hrs.	.....	0	0	0	17	13	6	Trace	
2[2]	White oak	50	Treated tree	7/27/51	3 hrs.	.....	.....	6	Trace	Trace	0	0	Trace	0
				7/27/51	6 hrs.	.....	.....	19	Trace	11	0	Trace	0	0
				7/27/51	9 hrs.	.....	.....	32	15	20	16	Trace	0	0
3	Cherry	34	Treated tree	6/27/52	3 hrs.	Trace	4	17	2	1	1			
	Cherry	24	Treated tree	6/27/52	3 hrs.	3	3	3	54	19	.....			
4	Cherry	22	Treated tree	6/28/52	14 hrs.	48	106	28	120	.....				
	Cherry	33	Treated tree	6/28/52	14 hrs.	2	97	52	88	155				
	Maple	43	Treated tree	6/28/52	14 hrs.	0	258	2	9	3	33			
5	Black oak	74	Treated tree	7/3/52	24 hrs.	11	22	31	1	13				
	White oak	78	Treated tree	7/3/52	24 hrs.	Trace	1,777	15	19	12				
	Maple	50	Treated tree	7/3/52	24 hrs.	33	87	9	3	.....				
	Hickory	82	Treated tree	7/3/52	24 hrs.	15	21	28	3	75				
	Cherry	70	Treated tree	7/3/52	24 hrs.	18	73	79	5	4				
6	Black oak	79	Treated tree	7/13/52	24 hrs.	74	63	10	120	0				
	White oak	78	Treated tree	7/13/52	24 hrs.	2	13	4	0	8				
	Maple	60	Treated tree	7/13/52	24 hrs.	183	13	.....	3	1				
	Hickory	82	Treated tree	7/13/52	24 hrs.	591	15	6	6	2				
	Cherry	69	Treated tree	7/13/52	24 hrs.	.....	76	4	7	6				
7	Black oak	67	Treated tree	7/27/52	16 hrs.	Trace	19	21	21	1				
	White oak	65	Treated tree	7/27/52	16 hrs.	6	148	29	12	14				
	Maple	50	Treated tree	7/27/52	16 hrs.	18	246	2	Trace	Trace				
	Hickory	75	Treated tree	7/27/52	16 hrs.	15	.....	3	Trace	4				
	Cherry	67	Treated tree	7/27/52	16 hrs.	28	47	23	28	40				

[1]A living tree, windthrown in winter 1950-51 with spiral grain, was half-girdled and arsenic applied to peeled side only.
[2]Two living windthrown trees: treated tree listed; control tree (untreated) no arsenic present.

However, penetration into the tree varies between species.

Anatomical examinations in connection with the above analyses reveal that tyloses are present in the vessels of all but the last formed annual ring of white oak. In cherry, gum plugs are present in the vessels of the heartwood. The vessels of black oak, hickory, and maple do not have these obstructions. Distinct zones of heartwood and sapwood are present in white oak, black oak, hickory, and cherry. In maple there is little or no heartwood.

White oak, black oak, cherry, and beech are killed in from three to eight days after treatment. Leaves of maple and hickory remain alive for as long as two months after treatment and isolated patches of living tissue varying in size are present. Living streaks occur, though sporadically, in all species, often starting from a point and broadening upward, sometimes causing incomplete bark separation.

Although arsenic is found below the treated girdles, closer examination reveals that the sporadically occurring streaks of dead tissue extend downward for short distances only. When trees are harvested six to seven months after treatment the stumps are still alive.

In trees treated during "sap peeling" season, resistance to peeling increases for as long as two to four weeks and is followed by loosening of the bark in white oak, black oak, maple and beech. In treated hickory, however, there seems to be no apparent change in peelability after treatment. In cherry, resistance to peeling increases and persists for as long as six months after treatment.

Anatomical studies reveal apparent shrinkage of undifferentiated, and differentiated cells of the tree's xylem, cells of the cambial zone, and cells of the phloem approximately six days after treatment, followed by a general radial collapse of these tissues in white oak, black oak, hickory, and maple (Fig. 3). While this behavior is also apparent in tissues of cherry, the frequently and regularly occurring multiseriate rays seem to resist radial-shrinkage, and phloem formed during the current year shrinks tangentially, pulling away from the rays (Fig. 4). In all species separation of bark from

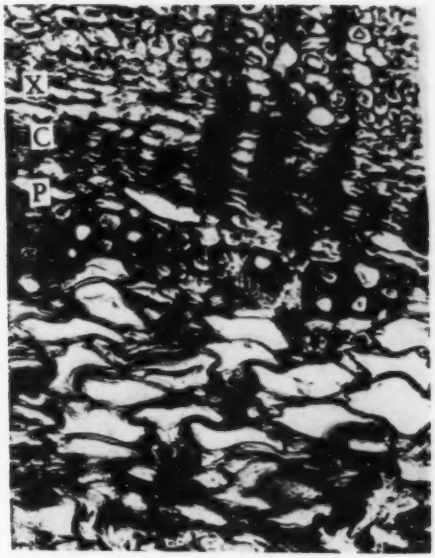


FIGURE 3. *Cross-section of black oak showing radial collapse of undifferentiated xylem, cambium and phloem.*

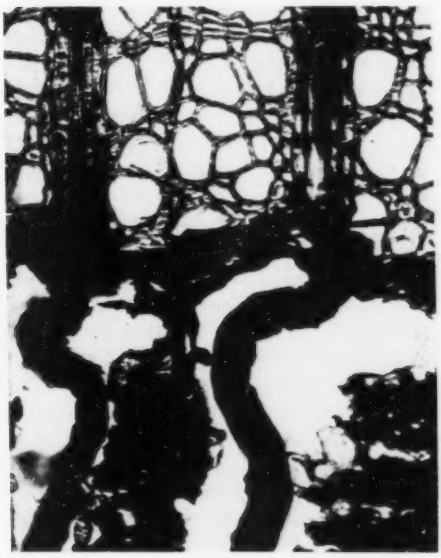


FIGURE 4. *Cross-section of black cherry showing both radial shrinkage of differentiating xylem, and cambium, and tangential shrinkage of phloem nine days after treatment.*

TABLE 3. Concentration of arsenic (ppm) in leaves and twigs at tops of trees.

	Tree No.	Height of tree (feet)	Concentration of arsenic (ppm)
White oak	1	50	27
	2	57	55
			75
	3	78	46
			32
			7
	4	78	23
	5	65	10
Black oak	6	74	14
	7	67	39
			6
Hickory	8	82	4
Maple	9	60	17
	10	43	7
			9
			Trace
	11	60	3
			3
	12	50	3
	13	60	55
	14	70	12
	15	69	14
	16	67	15
Beech	17	60	407

wood takes place in the incompletely differentiated, and differentiated, though immature, tissues of the xylem. In cherry, while this is also true, the rays persist as connective tissue between bark and wood, impeding peelability for as long as six months after treatment. The cells of the cambial zone degenerate until they cannot be readily recognized and the phloem is removed as part of the bark. Black oak and maple trees treated after the growing season show this same general pattern of tissue collapse. However, since the xylem is completely lignified and remains intact, subsequent delayed separation of bark from wood takes place in the cambial zone of both species. In cherry treated after the growing season, the rays retain the same characteristic resistance previously indicated in preventing separation between bark and wood.

Discussion

The constant association of bark slippage and xylem formation has been demonstrated and discussed by Bailey (1943), Brison (1933), and Schneider (1952). That bark slippage commences with the initiation of xylem formation and differentiation, and continues as long as new xylem tissue is formed and differentiating, and furthermore ceases when all the xylem tissue is completely matured, indicates not only a consistent occurrence of two phenomena simultaneously, but also an apparent dependence of bark slippage upon xylem formation. Although the initiation of phloem formation in cherry and maple takes place before and in hickory and ash subsequent to the inception of xylem formation, bark slippage is still consistently associated with xylem formation. The term "sap peeling," though commonly used to describe bark slippage, is misleading since it is in no way associated with sap flow. In maple trees the profuse exudation of sap from wounds incurred by the removal of specimens in midwinter without apparent increases in peelability belie the connotation of the term "sap peeling." The mucilaginous or gummy exudation from wounds in cherry beginning with the initiation of bark slippage in the spring and ceasing in late summer with the cessation of diametral growth is caused by increased metabolic activity of living cells in response to wounding and is not the cause of bark slippage. Bark slippage is rather an expression of an innate weakness of the distended walls of differentiating xylem cells before secondary wall formation, so that when bark is separated from wood during the growing season the break occurs in the zone of differentiating and immature xylary elements. Bailey (1943), in observing similar phenomena, declares that cellular proliferations obtained on the tangential surfaces exposed by peeling, splitting, tearing, or cutting originate from the immature xylem and not from the cambium.

Sodium arsenite applied to readily girdled trees during the growing season is rapidly translocated upward in the tree. There is a marked tendency for the solution to follow the longitudinal orientation of vascular elements of the xylem. For example, chemical analyses of specimens from a white oak (Table 2) girdled and treated on one side only reveals that the arsenic follows the spirally oriented xylem cells. The arsenic is present in the lower portion and absent in the upper portion of the tree on the treated side, above the girdle, and is absent in the lower part and present in the upper part on the untreated side.

Radial diffusion into the living bark occurs readily. A great number of analyses, not tabulated, bear this out. Centripetal diffusion, however, varies with wood structure. The bulk of the arsenical solution moves in the xylem closest to the cambial zone. This movement is apparently correlated with water transport and is generally in agreement with the findings of Rumbold (1920), who demonstrated that water transport in the ring-porous species is almost exclusively in the outermost annual ring. Richardson (1895) concluded from his studies of the effect of stem ringing in maple, beech, horsechestnut, oak, and laburnum that, in those species which form no heartwood, water ascends freely through the central portion of the stem as well as through the outer portion, and in those possessing heartwood the ascent of water is confined to the region of the sapwood. In general agreement were MacDougal and Overton (1927) in connection with sap flow and pressure studies in willow and walnut. MacDougal, Overton and Smith (1929) and MacDougal (1946) further noted that gums, tyloses, and gases obstruct sap flow in older tracheids and vessels.

In white oak where tyloses are present in vessels of all but the last formed growth ring, analyses reveal the radial diffusion of arsenic into the sapwood and for a short distance into the heartwood at the level of application and at sixteen feet above. In black oak, hickory, and maple, vessels of the sapwood and heartwood are without obstruction. Radial penetration is deeper into

the tree at the girdle. At higher elevations in black oak, hickory, and maple, analyses reveal not only deeper radial penetration, but in studies of hickory and maple suggest that more centripetally located tissues of the xylem are involved in upward translocation.

In cherry, the vessels of the sapwood are unobstructed, those of the heartwood are filled with gum plugs. While radial penetration at the level of application is deep into the heartwood, arsenic is limited to the sapwood twenty-five feet above the girdle. Upward translocation is apparently confined to the sapwood, and radial diffusion at that level is negligible. The tyloses of white oak and the gum plugs of cherry interfere with radial diffusion.

Whereas translocation is for the most part confined to the narrow sapwood in white oak, black oak, and cherry, in hickory and maple, with their considerably wider sapwood, there exists an opportunity for movement in deeper tissues away from the cambial zone and dilution of the arsenical solution. This perhaps explains the slowness with which maple and hickory are killed. The amount of arsenic translocated and the pattern of translocation is apparently dependent upon such anatomical features as physical obstructions in the vessels and relative amounts of heartwood and sapwood.

The greater movement of arsenic in the outer layers of wood strongly supports the feasibility of the practice of girdling to the wood and not into the sapwood immediately before applications of arsenite solution to the girdled face. Cuts into the sapwood of white oak may interfere with the upward translocation altogether or in other species may cause the arsenic to move upward in deeper tissues of the xylem away from the cambial zone, thus impeding the desired rapid killing effect. Girdling only to the sapwood results in the effective and uninterrupted translocation in the outer layers of the sapwood where the solution comes directly into contact with the cambial zone and adjacent living tissues.

Arsenic in its described movement through the tree quickly kills the cells of the cambial zone and surrounding living tissue. Upon treatment during the growing season tissue formation and differentiation end abruptly. Tissues are killed at a time when differentiating and differentiated xylem constitute the weak link between bark and wood. As these tissues shrink a stronger bond between bark and wood is formed, and bark slippage ceases for two to four weeks. It appears that subsequent shrinkage swelling, insect depredation, and fungi infestations cause the bark to separate from the wood. In cherry where resistance to peeling persists long after treatment, winter freezing and thawing may assist in bark removal. Trees with living leaves treated after the growing season are similarly killed. Since there is no zone of weakness between bark and wood not only is it difficult to make the girdle but those factors necessary for separation take longer to become effective. The prolonged tenacity with which bark adheres to the wood in cherry is caused by the apparent resistance of the rays to the factors responsible for separation.

Summary

This investigation was undertaken to study the nature of bark slippage and the role of sodium arsenite in bark removal from white oak, black oak, hickory, sugar maple, black cherry, and white ash trees.

Bark slippage is associated with xylem cell formation and differentiation. When bark is removed from wood the separation between the two is located within the differentiating and differentiated xylem cells.

Sodium arsenite applied to the peeled face of a girdled tree moves upward in the xylem adjacent to the cambial zone, quickly killing living tissue when the differentiating xylem constitutes a zone of weakness. Subsequent stresses brought about by shrinking and swelling of tissues, insect depredations, and fungi infestation cause the bark to separate from wood. Cherry reveals a prolonged resistance to peeling after treatment,

possibly due to distribution and structure of rays.

The movement of arsenical solution, as well as the rapidity of kill, apparently depend on wood structure, obstructions in the vessels and amount of heartwood and sapwood.

Literature Cited

ANONYMOUS. 1950. The chemical killing of trees to facilitate bark removal. Northeast Pulpwood Research Center, Durham, New Hampshire.

BAILEY, I. W. 1943. Some misleading terminologies in the literature of "plant tissue culture." Science 98: 539.

BENNETT, A. L. 1951. Planning chemical girdling for large scale pulpwood production. Paper presented to the Lake States Tech. Comm. Amer. Pulpwood Assoc., at Marinesco, Michigan. September 20.

BRISON, F. R. 1933. The storage and seasoning of pecan bud wood. Bull. Tex. Agric. Expt. Sta. 478.

EHRHART, N. O. 1951. Effect of chemical girdling on bark separation. Paper presented to the Committee on Coordination of Research of the Amer. Pulp and Paper Assoc., Madison, Wisconsin. May 16, 1951.

MACDOUGAL, D. T. 1946. Streaming movements of solutions in plants. Amer. J. Bot. 33 (5) 318-328.

MACDOUGAL, D. T. and J. B. OVERTON. 1927. Sap flow and pressure in trees. Science 65(1677): 189, 190.

MACDOUGAL, D. T., J. B. OVERTON, and G. M. SMITH. 1929. The hydrostatic-pneumatic system of certain trees: Movements of liquid and gases. Carnegie Inst. Wash. Publ. 397.

RICHARDSON, A. D. 1895. Effect of stem ringing on broad-leaved deciduous trees. Trans. Bot. Soc. Edinb. 20 (II): 337-339.

RUMBOLD, C. 1920. Effect on chestnuts of substances injected into their trunks. Amer. J. Bot. 7: 45-56.

SCHNEIDER, H. 1952. The phloem of the sweet orange tree trunk and seasonal production of xylem and phloem. Hildgardia 21: 331-366.

Measurement of Acorn Crops

BY
LESLIE W. GYSEL

DURING the past 10 years, measurements of acorn crops in different sections of eastern and central United States have been made by various wildlife agencies because of the importance of acorns as a food for animals. Foresters in Europe and the United States have also long been interested in the measurement of acorn crops and the subsequent development into seedlings. A variety of methods have been used to measure the size and to determine the quality of the crop. It is the purpose of this paper to summarize some of the factors affecting the production of acorns and to evaluate various devices and methods that have been used in the United States for acron crop measurements. Results obtained from recent studies of methods for measuring acorn production in Michigan are described. Seed measurement techniques presented may be applicable for the analysis of the seed crops of other heavy seeded tree species.

Factors Affecting Acorn Production on Trees

Before a review of crop measurement techniques, it will be of value to summarize the factors affecting acorn production, because the influence of the environment and individual variation in trees should be thoroughly considered in the planning and in the analysis of seed production studies.

Species

Although there is sometimes considerable variation in the production of acorns from individual trees of one species, a trend of production for a species may be ascertained for relatively small wooded areas (Downs and McQuilkin, 1944). Species differences are apparent in the data presented in Table 1; the black oak and white oak are dominant trees in a 10-acre woodlot and the red oak is in a separate woodlot in the same county. Conditions favoring good acorn production for one species in a specific area may not necessarily be good for another. Climate and other factors of environment may affect various species differently. Since little is known about the actual effects of most environmental factors on the seed production of trees, it is difficult to determine the size of area in which acorn production by species may have the same general trend. The production of red oak acorns for one year from trees of similar size, age, condition, and crown position varied in different stands located from 10 to 60 miles apart in southern Michigan. From north to south the relative production in each stand was as follows: poor, fair, good, poor. Acorn production of white oak in all of these stands was poor. Baker (1950), in summarizing German seed studies, concluded that in some years good crops are found over large areas while in other years good

The author is an associate professor and forest ecologist, Fisheries and Wildlife Department, Michigan State University, East Lansing. Journal Article 1901 from the Michigan Agricultural Experiment Station.

TABLE 1. Yearly acorn production by individual trees according to the average number of acorns per seed trap in two Michigan areas (surface area of trap—10.9 square feet).

Tree No.	Black oak (Rose Lake Exp. Sta.)				White oak (Rose Lake Exp. Sta.)				Red oak (Maple Rapids, Mich.)			
	1952	1953	1954	1955	1952	1953	1954	1955	1952	1953	1954	1955
1	1	18	51	69	1	0	22	5	0	1	18	46
2	8	27	101	102	0	0	0	0	0	1	3	38
3	7	21	53	44	2	0	12	2	0	17	11	38
4	1	2	19	12	0	0	7	1	0	7	1	1
5	2	27	67	57	1	0	23	0	0	4	6	15
6	2	7	6	19	1	0	11	5	2	1	3	0
7	23	26	29	65	3	0	0	9	1	1	2	12
8	1	11	61	58	1	1	23	0	1	30	33	97
9	2	25	77	38	2	0	23	23	1	17	22	42
10	10	49	75	79	2	0	70	5	0	21	22	113

crops may be local, largely due to the incidence of frost.

Characteristics of Individual Trees

Various investigators (Christinsen and Korschgen, 1955; Downs and McQuilkin, 1944; Wood, 1934) have indicated that individual oaks of the same species, similar in size, and under similar environmental conditions may vary considerably in the production of acorns. They also point out that some trees are habitually heavy or light producers. The variability of individual trees sampled in Michigan is indicated by the data presented in Table 1. Individual variations in acorn production in any one season and the seasonal variations make sampling of several trees for more than one season necessary to obtain an average or the range of production for an area.

Age

It is difficult to generalize about the relationship of the age of oaks and acorn production from the data now available. In studies of southeastern oaks, Downs and McQuilkin (1944) and Cypert (1951) found comparatively low acorn production below 14 inches d.b.h., varying rates of increase for some species to 20 inches or 30 inches d.b.h., and a leveling off or decrease in production over 30 inches d.b.h. In Missouri, Burns *et al.* (1954) found a weak relationship between acorn production and the diameter of five oak species 12 inches to 25 inches d.b.h. and from 30 to 80 years of age. They state that variation in seed production, probably due to heredity differences, almost completely obscured variation due to tree size and growth rate. Wood (1934) came to the same conclusions from seed studies with chestnut oak in New Jersey.

When bumper acorn crops occur, trees of all sizes may produce acorns. This occurred in northern Michigan in 1952. Kolesnichenko (1949) reports similar observations during years of high acorn production in southern Russia.

Climate

Little is known about the relationships of most of the factors of climate to acorn production; however, one climatic effect can often be seen — the killing of oak flowers as the result of low temperatures in the spring. This is a common occurrence in Michigan and has also been noted in West Virginia (Uhlig and Wilson, 1952).

There is apparently no definite periodicity in the occurrence of seed years, prob-

ably partly because of the variable effects of climate. Burns *et al.* (1954) found great variation from year to year in the abundance of acorn crops in Missouri. Kolesnichenko (1949) concluded that there is no definite periodicity either in occurrence of climatic factors or in the occurrence of seed years in southern Russia. Yearly acorn production in Germany is also quite variable according to crop measurements for 20 years presented by Schwappach (1895).

Crown Exposure

According to a study of acorn production in Michigan by Verme (1953), in which a detailed study of 33 trees was made, acorn production of a tree is greatest in the portion of the crown exposed to direct sunlight. Acorn production from the exposed crown areas was 11 times greater than the production of the lower unexposed crown areas for black oak and 5 times greater for white oak and red oak. These differences represent the greater production of the upper crown limbs. Acorn production of the outer parts of border trees was also larger than on the portions partly shaded by other trees. This resulted from a greater number of acorns per limb and a greater number of limbs on the exposed side of the tree.

It might be assumed that open-grown trees, which ordinarily have large crowns, produce large acorn crops. This is not always true because individual variations also occur in the open; some produce no acorns at all. Data obtained from Michigan experiments, however, indicate that open-grown trees that do produce acorns may have much larger crops per unit area of crown than any of the smaller crowned trees in closed stands of similar species and age (Table 2).

As a result of the light requirements of oak for acorn production, it is also logical to assume that intermediate and suppressed trees in forest stands would generally be low producers. Moody (1953) found that this relationship existed for oaks in Louisiana, and Kittredge and Chittenden (1929) made similar observations in northern Michigan.

From acorn counts made according to aspect, Verme (1953) found that the south and west facing sides of tree crowns exposed to full sunlight were more productive than those facing north or east.

Devices and Methods for Measuring and Analyzing Acorn Crops

Tree Counts

The most accurate determination of the number of acorns on a single tree can be made by a complete count before the acorns fall or are heavily utilized by animals. This can be done rather easily on small trees; on large trees, however, complete counts are difficult and time consuming. Special climbing apparatus is needed for tall trees in closed stands. Except for special individual tree studies, there appears to be little need for complete counts. In years of high acorn production, counts on sample crown areas can be made instead of complete counts. Verme (1953) found close agreement between sample area counts and complete counts. To determine annual fluctuation in acorn abundance in stands of stunted post oak in East Texas, Petrides *et al.* (1953) suggested counting the acorns occurring on the first twenty-five twigs on each tree. Field

TABLE 2. Production of acorns in 1955 from trees in different exposure conditions at the Rose Lake Wildlife Experiment Station, Michigan.

Position of trees	Average number of acorns per seed trap	
	Black oak	White oak
Interior of closed stand	55	6
Border of closed stand	92	
Open field	95	53

Basis: Interior of closed stand—20 trees of each species; border of closed stand—6 trees; open field white oak—5 trees; black oak—4 trees. Surface area of seed traps—10.9 square feet.

glasses and spotting scopes may be used to make sample counts on larger trees.

Ground Counts

A simple method to obtain an estimate of the acorn production of individual trees is to count the acorns on the ground. Dalke (1953) describes a study of mast yields acre plots established at one-chain intervals along 5.6 miles of surveyed lines. Periodic collections for studies of the acorn yields have also been made from mil-acre quadrats used in connection with seed traps (Downs and McQuilkin, 1944). For larger samples, acorns have been collected from triangular sample plots consisting of approximately 10 percent of the crown area (Cypert and Webster, 1948; Allen and McGinley, 1947).

The size of the sample that can be used depends upon the accuracy required, the time available for field collections, and the density of the trees. A large triangular sample would be difficult and laborious to use in a closed stand.

Seed Traps

Different kinds of devices for catching seeds have been devised. They serve to concentrate the seed in a small area to facilitate the analysis and lessen acorn losses caused by animal use. One type consists of a shallow box, usually one-quarter mil-acre in area with wood or wire sides and bottom, and poultry netting covering the top. (Cypert, 1951; Downs and McQuilkin, 1944; Burns *et al.*, 1954). Another type has the form of a funnel or inverted pyramid, having a base 3.3 feet on a side. Easley and Chaiken (1951) describe a trap of this type constructed of corrugated kraft paperboard which was tacked to a light wooden frame and supported by four wooden legs. A wire basket was attached to the bottom opening to hold the seeds. For a long-term acorn-production study in Michigan, a modification of this trap was made; canvas treated with waterproofing material was used in place of the paperboard, and welded ⅛-inch iron or galvanized pipe was used for the frame (Figure 1).

Before the Michigan acorn study was started, traps of different designs were tested in the field. The shallow-box type, if solidly constructed, was heavy and difficult to carry in the woods. The cost was approximately $7 to $9 per unit. The funnel types were light and portable. The canvas unit with a light iron frame was the easiest to handle in the field and the most durable. Traps of this type used in Michigan for five seasons showed very little wear. Kraft paperboard traps were not as easy to handle in the woods. After five collecting periods of use, the paper traps were still usable in southern Michigan; in northern Michigan, however, traps of this type which were left in the field were not usable after three years. Canvas traps with iron frames cost approximately $5 to $6 per unit and the paperboard units with wooden frames approximately $3 to $4.

Tests to determine the efficiency of the various types of seed traps were made. In one experiment 1,000 black oak and white oak acorns were dropped from a height of 30 feet on five types of traps. As indicated in Table 3, losses of the acorns by deflection are large enough to merit consideration in evaluating estimates from seed traps. Numbers of acorns deflected from uncovered traps were large enough to make the data useless unless a correction factor was used. A vertical collar around the

TABLE 3. Seed trap tests.

Type of seed trap	Acorns deflected from traps	
	White Oak	Black Oak
	Percent	Percent
Canvas funnel-type with 1-inch-mesh poultry netting	11	11
Canvas funnel-type uncovered	32	12
Paperboard funnel-type with 1-inch-mesh poultry netting	17	8
Paperboard funnel-type uncovered	78	58
Rectangular box-type with 1-inch-mesh poultry netting	15	7

Basis: 100 acorns of each species dropped on each trap from a height of 30 feet.

FIGURE 1. *A funnel-type seed trap made of canvas. The frame is made of ⅛-inch iron pipe welded at the corners, and the stakes are wood. This trap has been efficient and durable.*

traps above the netting would probably reduce loss of acorns due to deflection. This was not tested, however.

A series of tests with marked acorns indicated less loss due to small rodents in the funnel-type traps than in the rectangular box traps. In general, acorn losses from the funnel type traps were small, although during the latter part of the period of acorn fall in Michigan, some acorns were usually taken. Various shapes of wire baskets for catching acorns were tried for the purpose of eliminating the loss, but these were of little value. Moth balls were tried as a repellent, but were not effective either.

Location of Sampling Units

To determine the acorn production of a stand, seed traps or ground quadrats may be set at random throughout a stand. A grid system could be used for establishing the location of the sampling units. It is also possible to obtain similar data by placing the sampling units under randomally-selected trees and expanding the data on the basis of a stand table. Although a statistical comparison of the two methods was not made, it appears that the latter may be more efficient because each trap or quadrat is placed under the crown of a tree. With a random placement of the sampling units using a grid system, some would be a sample of only small portions of tree crowns.

In the sampling of individual trees by seed traps or open quadrats, the sampling units may be placed at random under any portion of the crown or under a selected area. In a Michigan acorn production study, seed traps and quadrats were placed only under the portions of the crown exposed to light because the majority of acorns were produced in that area. This method can be used best when sampling is done with one or two small traps or quadrats per tree.

Analysis of Field Collections

The kind of tests and observations to be used in analyzing the acorn samples depends largely on the objectives of the seed study. The simplest breakdown would be in two categories, sound and damaged. From an analysis of the damaged acorns, it is possible to determine the proportion affected by insects, animals, and sometimes by fungi or bacteria. It is not always possible, however, to determine in the field whether a malformation of the cotyledon and embryo is due to fungi. In the Michigan acorn study, only a part of the damaged acorns originally designated as infected by fungi in the field produced fungi in culture; other malformations were apparently due to other kinds of physiological changes. It is also possible to make counts of acorns damaged by animals, although sometimes this can be only approximate because only small pieces of acorns are left. The proportion of small or aborted acorns can be determined easily.

Within the damaged category, further indication of the amount of damage may be of value in some studies. Partly damaged acorns may actually furnish much wildlife food and in the case of white oak, sprouting will occur even though a part of the cotyledon is damaged. In Michigan

studies two degrees of damage were recognized, one category included acorns less than 50 percent damaged and a second category more than 50 percent damaged.

Losses of acorns on the ground due to animals may be estimated by comparing a total count in the seed trap with the count made from adjacent open quadrats (Downs and McQuilkin, 1944). A periodic tally of marked acorns will provide an indication of the rate of removal.

The amount of an acorn crop removed and damaged by the animals in the trees can only be approximated. Cypert and Webster (1948) obtained this kind of data by direct observations; however, only rough approximations could ordinarily be obtained in this way. It is often difficult to follow the actions of animals in trees, even with powerful glasses; also, a large portion of the removals may be made at night. Losses from the trees may also be indicated by empty cups on the ground. Korstian (1927) determined animal losses by subtracting the total acorn count of sample areas from the total count of cups. There may be a considerable error in this method because the cups of some species do not fall for many months. Also, field observations in Michigan indicated that both the acorns and the cups are often removed by animals.

Use of Sample Data

The size of acorn crops determined by a sampling method may be designated in several ways. For comparing the relative size of crops on two or more areas, the average number of acorns per seed trap or open quadrat may be used. If the crown areas of the sample trees are measured, the trap or quadrat data can be expanded according to the ratio of the crown area to the trap or quadrat area. If the number of trees per acre is known, the total number of acorns can be determined from the sample tree data. In addition to number of seeds, it is often desirable to express acorn production in terms of weight, either "oven-dry" or "as collected." The number of acorns per unit of weight can be easily obtained by weighing small samples of known numbers of acorns during the collecting season.

Analysis of Sampling Methods

Number of Seed Traps per Tree

In the Michigan acorn studies, two seed traps were placed under the fully developed portion of the crown exposed to sunlight, generally on opposite sides of the tree. During the field collections it was noted that differences in the number of acorns in each of the paired traps were relatively small. An analysis of 45 trap pairs was made by comparing the sum of the acorns in a pair with the number of acorns in one of the traps doubled. For 30 of the 45 pairs contrasted in this way, the difference between traps was 15 percent or less, and for 39 of the 45 pairs the difference was 25 percent or less. In a separate sampling study in which there were three and four traps per tree, 10 of 14 traps compared in the same manner had a difference of 15 percent or less and 13 had a difference of 25 percent or less.

Differences between trap pairs indicated variations in the intensity of acorn production within the exposed crowns of individual trees. During a study of acorn production on individual trees, it was found that differences sometimes occurred on adjacent branches. A separate study of 50 trap pairs was made, part of which were samples from the north and south facing sections of tree crowns and part from east and west facing sections. No consistent relationship between acorn production and aspect of the crown could be determined from these samples.

The number of sampling units per tree depends upon the accuracy desired in the study. Many seed traps or other sampling units could be used to measure possible variations that might occur in different parts of the crown. However, both the traps and the time to service them are costly. This cost would have to be balanced against the funds and time available

for the entire study. It is possible that one or two small sampling units per tree may be adequate for many studies.

A Comparison of Seed Trap and Open Quadrat Samples

During the weekly collection of data for the Michigan acorn study it was noted that the number of acorns from the seed traps was generally very similar to the numbers collected from the adjacent open quadrats. To make a comparison, total yearly counts of the average size acorns were analyzed from three different areas, each having twenty seed traps and twenty open quadrats placed under the exposed part of the crown of ten to fifteen individual trees of similar size and crown position of each species. It was found that the differences between samples collected in the seed traps and the adjacent open quadrats were small in comparison to the total number in the samples. Analyses of variance for testing mean differences of trap and quadrat data, with the assumption that both represented random samples, indicated no significant difference at the five percent level (Table 4). Tests of the average difference between paired traps and quadrats also indicated no significant difference except for one of the 10 sample groups.

It might be assumed that ordinarily the count in the ground quadrats would be noticeably less than that in the traps due to removal of acorns by animals. Where the ground rodent population was especially high as in the Rose Lake area (Table 4), the trap counts were often slightly higher than the open quadrat counts. However, the losses in these small sample units between weekly collections in this area and also in the other areas during the time when most of the acorns were falling were relatively small. In contrast, the total removal of acorns throughout the entire fall season on the small sample units was generally large; often all of the sound acorns were completely gone by December.

A tally of the acorns in the seed traps or quadrats may be made at any interval. The longer the periods between the collection of seeds, however, the greater the possibility of losses from the traps or quadrats. In some areas the continuous removal of acorns by small rodents, especially during the latter part of the collecting season, may cause significant errors. In addition, if the traps are not serviced at regular intervals, leaves and twigs may cover the netting so that acorns remain on the surface of the trap readily available to many animals.

TABLE 4. Means of the total number of average size acorns obtained from seed traps and adjacent open quadrats in three Michigan areas.

Area	Species	Year of collection	Means of sample counts: Seed trap	Means of sample counts: Open quadrat	Standard error of mean[1]
Rose Lake	Black oak	1953	20.9	12.8	2.8
Exp. Sta.	Black oak	1954	50.8	44.7	6.2
Do	Black oak	1955	54.8	53.0	5.8
Do	White oak	1954	16.5	18.4	3.6
Flat River					
Game Area	Black oak	1953	29.1	30.1	3.4
Do	Black oak	1954	35.9	39.2	4.1
Do	Black oak	1955	40.7	34.3	4.9
Maple Rapids	Red oak	1953	9.7	8.4	2.1
Do	Red oak	1954	11.3	12.0	2.4
Do	Red oak	1955	30.3	40.1	7.2

[1]Computed from the pooled estimate of variance. Basis: 10 or 15 sample trees in each area with 20 traps and 20 open quadrats. Surface area of seed traps—10.9 square feet.

Acorn counts from the seed traps may be less than the total number falling in the traps due to deflection as indicated in Table 3. A correction factor can be applied to the trap counts to compensate for this error. This factor varies according to the type of trap and the distance the acorns fall. For the trap data presented in Table 4, which was obtained from covered funnel-type canvas traps at an approximate distance of 30 feet from the bottom of the crowns, this factor would be 1.11.

Number of Sample Trees

In Michigan 15 dominant and codominant oaks of each species in even-aged stands

were sampled for a study designed to compare the general productivity of different stands. An analysis of the variability of individual trees indicated the necessity of at least a sample of this size. The standard error of the analyzed data ranged from 10 percent to 25 percent of the means (Table 4). To decrease the standard error considerably it would be necessary to increase the number of sample trees several times. In one area, to reduce the standard error to 5 percent of the mean, 40 sample trees would have been required, and in another area 100 trees.

Summary and Conclusions

1. Acorn production may vary according to the species and to some extent to age of the trees within a stand.

2. There is considerable variation in acorn production among individual trees of a species under similar environmental conditions.

3. The climate of an area has an effect on acorn production although the effects are apparently not cyclic.

4. The intensity of acorn production of a tree is much greater in the portion of the crown exposed to full sunlight than in the unexposed portion of the crown.

5. The measurement of acorn crops by counts of acorns on the trees can be made by complete or sample tallies. Field glasses or spotting scopes can be used to make the counts. This method is best adapted to studies of individual trees.

6. Estimates of the acorn production of trees can be made from periodic collections from sample areas on the ground beneath the trees.

7. Seed traps can be used to facilitate the collection and analysis of acorn samples and to lessen losses due to animals. Some of the acorns are generally deflected from the traps. A correction factor can be used to compensate for this loss.

8. Differences in the acorn count between pairs of seed traps and also larger groups of traps under the same exposed tree crown were relatively small. One or two sampling units per tree may be adequate for many studies.

9. Differences between samples collected in seed traps and adjacent open quadrats were generally small in comparison to the total number of acorns in the sample. An estimate of the acorn production of a tree approximately equal to that from seed traps may be obtained from weekly collections of acorns from ground quadrat samples.

10. To obtain an adequate estimate of the acorn production of the trees of one species similar in size and position in the stand, samples from at least 15 trees will be required.

Literature Cited

ALLEN, D. L. and H. D. MCGINLEY. 1947. A method for the year to year measurement of mast yields. J. Wildlife Manag. 11: 184-185.

BAKER, F. S. 1950. Principles of silviculture. McGraw-Hill Book Co. 414 pp.

BURNS, P. Y., D. M. CHRISTINSEN and J. M. NICHOLS. 1954. Acorn production in the Missouri Ozarks. Univ. of Missouri Agric. Exp. Sta. Bull. 611. 8 pp.

CHRISTINSEN, D. M. and L. J. KORSCHGEN. 1955. Acorn yields and wildlife usage in Missouri. Trans. 20th N. A. Wildlife Conf. 337-357.

CYPERT, E. 1951. Suggestions for the management of oak forests for mast production. Mimeo. paper presented at the S. E. Assoc. Game and Fish Commissioners Conf. Edgewater Park, Miss. 8 pp.

CYPERT, E. and B. S. WEBSTER. 1948. Yield and use by wildlife of acorns of water and willow oaks. J. Wildlife Manag. 12: 227-231.

DALKE, P. D. Yields of seeds and mast in second growth hardwood forest, South-central Missouri. J. Wildlife Manag. 17: 378-380.

Downs, A. A., and W. E. McQuilkin. 1944. Seed production of southern Appalachian oaks. J. For. 42: 913-920.

Easley, L. T., and L. E. Chaiken. 1951. An expendable seed trap. J. For. 49: 652-653.

Kittredge, J. and A. K. Chittenden. 1929. Oak forests of northern Michigan. Mich. Agr. Exp. Sta. Spec. Bull. 190. 47 pp.

Kolesnichenko, M. V. 1949. Poldonoshinie dooba (The fruition of oak). Lesnoye Khoziastivo (U.S.S.R.) 7: 26-28.

Korstian, C. F. 1927. Factors controlling germination and early survival in oaks. Yale Univ. School For. Bull. 19. 115 pp.

Moody, R. D. 1953. Mast production of certain oak species in Louisiana. Mimeo. paper presented at the S. E. Assoc. Game and Fish Commissioners Conf. Chattanooga, Tenn. 26 pp.

Petrides, G. A., P. Parmalee, and J. E. Wood. 1953. Acorn production in East Texas. J. Wildlife Manag. 17: 380-382.

Schwappach, A. 1895. Du Samenproduction des wichtigsten Waldholzarten in Preussen. Z. f. Forst. vu. Jagdwes. 147-174.

Uhlig, H. G. and H. L. Wilson. 1952. A method of evaluating an annual mast index. J. Wildlife Manag. 16: 338-343.

Verme, L. J. 1953. Production and utilization of acorns in Clinton county, Michigan. Unpub. M.S. Thesis. Mich. State Univ. 77 pp.

Wood, O. M. 1934. A brief record of seed productivity for chestnut oak in southern New Jersey. J. For. 32: 1014-16.

A Spherical Densiometer For Estimating Forest Overstory Density

BY
PAUL E. LEMMON

FORESTERS, ecologists, range scientists and many others recognize that forest overstory is important to the plant community. It affects such basic habitat factors as light, moisture, wind and temperature.

Light is required for the growth of most plants and is characterized by four attributes: quality, direction, intensity, and duration (Weaver and Clements, 1929). When a forest overstory is present it may alter one or more attributes of the light that penetrates into lower portions of the tree crowns, to the understory plants, and to the forest floor.

A forest overstory may intercept snow and rain causing a loss of potential soil moisture due to evaporation from exposed and greatly increased surface areas. It may have an important influence upon the accumulation and melting of snow and ice (Craddock, 1954).

Interception of rainfall by a forest overstory and increased evaporation result in a cooling effect on the temperature. Insulation against direct radiation from the sun, moderation of wind currents, and cooling due to shading, reduce transpiration from plants and evaporation from both soil and plants.

Many instruments and methods have been used to study the influences of forest overstory. Examples of such instruments and methods are: photometers (Weaver and Clements, 1929; Matusz, 1953), light meters (Jackson and Harper, 1955), photographic methods (Suzuki and Satoo, 1955), densiometers and ceptometers (Robinson, 1947; Ingebo, 1955), vertical crown projection methods, and ocular estimations of overstory density.

Studies of forest overstory density are made by foresters to establish spacing standards in forest thinnings and to determine light requirements for regeneration. Soil Conservation Service technicians record measurements of overstory density during field studies made to relate soil and other environmental factors to growth of trees and grasses. The author designed an instrument to make measurements of overstory density easily and accurately. This paper describes the instrument and gives some data on its reliability in use.

Development and Description of Instrument

Ocular estimations of overstory density were used in preliminary work. These were

The author is Soil Scientist (Forestry), U. S. Department of Agriculture, Soil Conservation Service, Portland, Oregon. Acknowledgment is made to the following for their help and encouragement in developing the instrument, testing it and in preparation of this manuscript: Clyde E. Deardorff, Soil Scientist and Orlo W. Krauter, Woodland Conservationist (West), Portland, Oregon; William M. Johnson, Principal Soil Correlator, Berkeley, California; and many other Soil Conservation Service field personnel throughout the West.

intended to estimate relative area of crown coverage by vertical crown projection methods. The first instrument used by the author was a densiometer patterned after that described by Robinson (1947). A flat mirror was used in this instrument which limited the size of the overstory sample being measured. A large number of measurements was therefore needed to get a reliable estimation. The instrument was also large and inconvenient to carry. Study and discussion of the problem suggested the use of a curved or spherical mirror to reflect a larger segment of the overstory. Ingebo (1955) employed this principle in his ceptometer. An improved method of delineating an area of overstory on the mirror for density estimations was sought. After much study and trial, spherical densiometers described briefly in a discussion of Ingebo's paper were devised.[1]

Two models, A (Fig. 1) and B (Fig. 2), have been adopted as standard. Each employs a highly polished chrome mirror 2½ inches in diameter and having the curvature of a 6-inch sphere. The convex side of the mirror is used in Model A and the concave side in Model B. Each has some advantages over the other. Concave mirrors invert the reflected image. The reflection is less clear than in convex mirrors unless the viewer closes one eye. The area of overstory being measured by a concave mirror can be more nearly overhead because the head of the viewer may be easily kept outside the angle of reflection. Identification of reflected objects is more difficult in the concave mirror due mainly to image inversion.

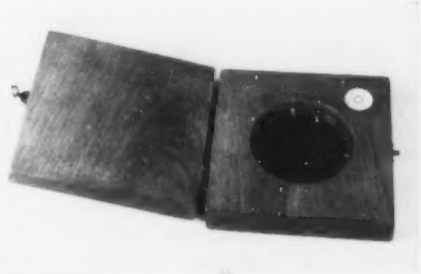


FIGURE 1. *Spherical densiometer, Model A, with estimating grid scratched on the surface of the convex mirror.*

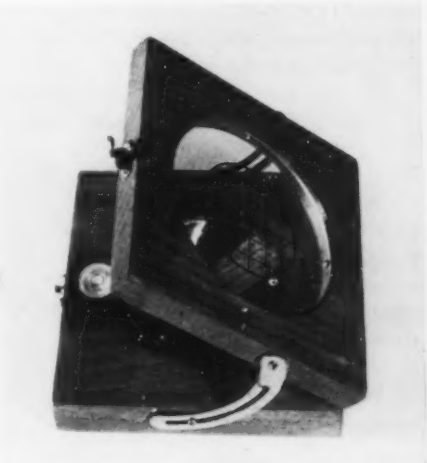


FIGURE 2. *Spherical densiometer, Model B, with estimating grid superimposed between the eye and the surface of the concave mirror.*

[1]The first spherical densiometer was a 6-inch hemisphere with a mirror surface and a flange at the base like the brim of a hat. It was fixed to a camera tripod. Many kinds of grids for estimating area were tested. One was a wire hoop with square cross-sectional markers held about one to two feet above the hemisphere in such a way that its reflection fell precisely on a circle marked on the surface of the mirror. This grid could be superimposed at will on the mirror at any point representing changes in direction of incoming light through the overstory. This grid is perhaps the most accurate and has several interesting possibilities. However, there are some mechanical problems of support and the instrument is cumbersome. In fact, the hemispherical mirror itself is larger than necessary because it reflects side and even ground areas as well as overstory. Only a small part of a spherical mirror is needed to reflect a large enough area of overstory for measurement.

TABLE 1. Analysis of variance: test of spherical densiometer.

Source of variation	Degrees of freedom	Sums of squares	Mean squares	Variance ratios (f)
Instruments	1	480	480	3.38
Forests	6	63,792	10,632	74.87**
Operators	3	308	103	0.73
Interactions:				
Instruments and forests	6	1,917	320	2.25
Forests and operators	18	3,302	183	1.29
Instruments & operators	3	346	115	0.81
Error	18	2,552	142	
Total	55	72,351		

**Significant at 99% level.

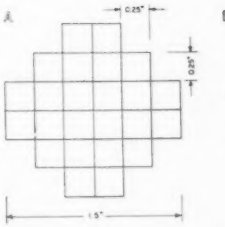
The mirrors are mounted in small wooden recessed boxes with hinged lids similar to compass boxes. The over-all dimensions are about $3\frac{1}{2} \times 3\frac{1}{2} \times 1\frac{1}{8}$ inches. A circular spirit level is mounted (recessed) beside the mirrors. Positive slide fasteners are provided in Model B which allows the lid to open to an angle of about 45 degrees.

Cross-shaped and circular grids with squares and dots are used to estimate overstory coverage by tree crowns. Grids are of two kinds: (1) those scratched upon the surface of the mirror, Model A, and (2) those superimposed between the mirror and the eye, Model B.[2]

The cross-shaped grid scratched upon the convex surface of the mirror in Model A has 24 quarter-inch squares (Fig. 3A). Instructions for using the densiometer and cumulative values for the squares on the grid are shown on a chart that is attached to the inside of the box lid (Fig. 3B). It is easier and faster to estimate the relative amount of overstory coverage with this instrument by assuming the presence of 4 equi-spaced dots in each square and by counting dots representing openings in the canopy. The percentage of overstory density is then assumed to be the complement of this number. Each assumed dot is assigned a value of one percent in this case. A slight discrepancy exists between estimations using the squares and estimations by counting assumed dots, because there are only 96 dots in the entire grid area. Cumulative values of the squares shown in the chart add up to 100 percent for the entire area within the grid. If desired, one may calculate the exact percentage values for each assumed dot and thereby make the two methods of use exactly comparable. Data presented for this instrument were based on cumulative values of the grid squares.

Model B has a circular grid. The circle is one and one-half inches in diameter superimposed over quarter-inch squares. Each square has four equi-spaced dots (Fig. 4A). This grid is made from a positive print of a photographic film mounted between thin sheets of plexiglass and fitted into the window of the box lid. Instructions for operating Model B are given on a chart mounted on the bottom of the instrument box (Fig. 4B). The operator estimates overstory density by counting the dots representing overstory openings and assuming this to represent the percentage of non-covered overstory area. Here again a slight discrepancy exists because there are only 96 dots included within the area of the circular grid. Exact percentage values for each dot may be calculated to estimate the entire circular area as 100 percent. This

[2]Grids superimposed between the mirror and the overstory have been used by Ingebo, 1955 and Robinson, 1947, and were tried by the author. (See footnote 1).



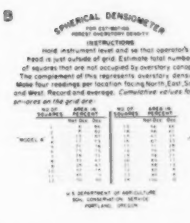


FIGURE 3. A. *Cross-shaptd grid scratched on the convex surface of the mirror in Model A. Each square is ¼ inch o na side.* B. *Instructions for using Model A. This is fastened to the inside of the lid of the mounting box.*

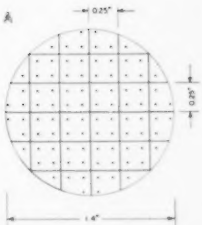


FIGURE 4. A. *Circular grid superimposed between the eye and the concave mirror in Model B. Each square is ¼ inch on a side.* B. *Instructions for using Model B. This is fastened to the bottom of the mounting box.*

refinement is not considered necessary for ordinary use of the instrument.

The kind, size, and shape of the grid may be designed to meet the needs. Such things as amount of lateral coverage, desire for sampling specific overstory areas from a point, and ease in making the estimation are considerations in designing the grid. Small aberrations in the cross-shaped grid scratched on the surface of the curved mirror are thought to be within the accuracy of the instrument and are not important sources of error.

The degree of curvature of the mirror may be selected within limits to meet the needs of the investigator. It seems important, however, to have mirrors with known physical properties of curvature. Mirrors with curvature of a 6-inch sphere have been satisfactory in work with Douglas fir, ponderosa pine, lodgepole pine and other western conifers. They reflect an area of overstory large enough for accurate estimations of covered and non-covered areas. Curvature has not caused appreciable inaccuracy due to scratching a grid on the surface, reading the instrument, or fitting it into small compact portable units. Standardization of mirror curvature, grid size and design and methods of using the instrument are necessary to provide comparable information that can be duplicated. Operators need a little training to become consistent in the use of the instrument. Judgment and experience is needed to differentiate between overstory areas that are considered completely covered by the overstory and those that have thin but uniformly distributed coverage. In the latter case it may be necessary to estimate the area of many small irregular openings and reduce the percentage overstory density by the sum of these. Training and experience are needed for each different forest species or type because of the differences in overstory characteristics. The season of the year is important when making measurements in forests containing deciduous species.

Experience has shown that sufficient accuracy can be attained with the spherical densiometer by holding it as nearly level as possible in the hand. This is made possible by installing a circular spirit level in the mounting box. No mechanical support, such as a tripod, is needed. This adds to the practicability of the instrument in use.

Using the spherical densiometer, Soil Conservation Service technicians measure overstory density facing each of the four cardinal directions at each sample point. Since the overstory area measured by the spherical densiometer is not directly overhead, this gives added information about overstory density in different directions. Slope percentage and aspect are always measured by supplementary equipment. It

has not been necessary to install compasses in the densiometers currently in use although this could easily be done if desired.

Variations in Overstory Density

Overstory density measurements were made in several ponderosa pine forests in south central Oregon and south central Washington. Each of four different operators used both Model A and B instruments. Each operator measured the overstory density at four points, north, east, south, and west around a reference tree. The reference tree in each case represented a typical dominant or codominant in the stand. The points selected around each reference tree were far enough away so that the crown of the reference tree was just outside the overstory area being estimated. This is equivalent to 28 forest measurements made by each of four different operators by each of two different instruments. Care was taken to prevent one operator from knowing the results of any previous measurement during the work.

The data were subjected to an analysis of variance by standard procedures (Table 1). There proved to be no significant difference among measurements made by different operators or with different instruments. None of the interactions were significant. The only variable showing significance was that of forests. All other sources of variation were therefore pooled. When the error term was thus increased to 49 degrees of freedom, the variance ratio due to forests was 60.9, a value significant at the 99 percent level.

Fiducial limits of the mean of total means were set up for different levels of probability based on this latter analysis. These fiducial limits were based on means of four measurements.[3] Division by four would show expected variations at the different probability levels for actual overstory density measurements. For instance, the overstory in forest 42 (instrument A, direction N and instrument B, direction N) was measured 8 times with the following results: 71, 71, 67, 67, 71, 65, 74, and 66. The mean of these measurements is 69 percent. At the 70 percent level of probability results are reliable within ± 1.3 percent—a coefficient of variation of 2 percent of the mean. At the 95 percent level of probability the results are reliable within ± 2.4 percent—a coefficient of variation of a little over 3 percent. At the 99 percent level of probability the results are reliable within ± 3.1 percent—a coefficient of variation of a little over 4 percent. Both instruments apparently give consistently accurate measurements of forest overstory density, regardless of operator. The spherical densiometer is therefore a practical and reliable instrument for obtaining comparative information about forest overstory density for whatever purpose this information may be sought.

In the past, overstory density usually has been estimated and assigned to classes, both in accumulating field information and in analyzing results. With the spherical densiometer this is not necessary, for overstory density can be measured easily, quickly and accurately. If overstory density classes are needed to simplify calculations and analyses the class interval can be more narrowly defined by data obtained with this instrument.

Tests have been made to show reliability when forests are assigned to 5 percent overstory density classes by the spherical densiometer. Data used in computing Table 1 have been combined with additional data for this test (Fig. 5). Each of the 416 single measurements was allocated to a density class established by the average of the 4 measurements of each individual forest overstory sample. These included measurements by four different operators. For instance, forest 42, instrument B, direction N; the measurement 71 by operator 1 was placed into density class 66 to 70 established for the average (69) of 71, 65, 74, and 66, the measurements of operators

[3]The limits are ± 5.3 at the 70% level, ± 9.4 at the 5% level, and ± 12.5 at the 1% level.

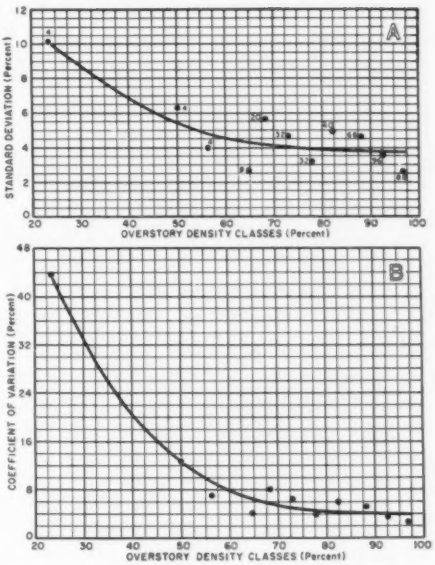


FIGURE 5. *Relation of standard deviation and variation coefficient to overstory density classes.*

1, 2, 3, and 4, respectively. Variation shown in Fig. 5 has been calculated around the mean of all the measurements within each class and not from the central class value. The data include all 224 single measurements used in Table 1 and 192 additional measurements all made with instruments of the Model A type. These additional measurements were equally divided among four different operators and included, as before, readings facing N, E, S, and W at each point in 9 additional forests. These forests included coast Douglas fir in western Washington, ponderosa pine and mixed coniferous forests in northeastern Washington.

The standard deviation and coefficient of variation of the mean of all measurements within each class were plotted against the mean (Fig. 5). Harmonized, freehand curves were drawn among the points. These curves indicate that variation among measurements increases as the overstory density decreases. This seems to agree with the findings of Jackson and Harper (1955). There is little significant change in consistency of measurements above about 60 percent overstory density. Actually, the reliability of measurements in forests of only 50 percent overstory density is entirely satisfactory for most purposes. In use, the reliability would be reflected by that portion of the curves to the right of the 50 percent overstory density class because an operator will naturally tally the areas that are covered with overstory instead of the areas that are not covered when measuring forests of low overstory density.

When assigning forest overstory to density classes by means of the spherical densiometer some loss in accuracy results. If 5 percent classes are used as in Fig. 5A, reliability of measurements of about the order of $\pm$ 5 percent can be expected (with a probability at about the 68 percent level) for all density classes above 50 percent. This is in contrast to a reliability of about $\pm$ 1.3 percent when actual measurements with the spherical densiometer are tabulated. The coefficient of variation (Fig. 5B,) amounts to about 5 percent for all density classes above 70 percent and increases to about 13 percent for classes down to 50 percent overstory density.

Summary

The spherical densiometer has been designed as a pocket type, easily used, highly accurate and practical instrument for determining from a point the relative amount of light that is cut off by specific areas of the forest overstory. It can be used equally well by the scientist doing highly technical research work, or by the practicing forester, range conservationist or plant ecologist.

Two models of the spherical densiometer, A and B, have been standardized. Both are based on the use of curved (spherical) mirrors which reflect the overstory conditions at a point and make possible the estimation of relative amounts of area covered and not covered. Measurements

are made by means of various kinds of grids. Model A has a cross-shaped grid with ¼-inch squares scratched upon the surface of the mirror. Model B is provided with a circular grid superimposed over ¼-inch squares, each of which contains 4 equispaced dots. This is mounted in the lid of the mounting box to be superimposed between the mirror and the eye of the operator.

Trials by different operators with both instruments have shown that there is no significant difference among measurements of overstory density made by different operators or with different instruments. Differences in forest overstory density, however, can be assessed with an unusually high degree of fidelity. For instance, at the probability levels of 70, 95 and 99 percent, average measurements of the same overstory area can be expected to be within ± 1.3, ± 2.4 and ± 3.1 percent respectively. These variations are about 2, 3, and 4 percent of the average overstory density measurements respectively (the coefficients of variation.)

Tests were made to show the influence upon expected variability of measurement when assigning overstory density to class. Two conclusions are obvious: (1) Variation among replicated measurements increases with a decrease in the overstory density. This is important in forests with overstory density varying from 100 down to about 50 percent. In forests with lower overstory density the operator naturally tallys the area covered with overstory instead of the area not covered. The variation in reliability, therefore, never exceeds that shown for about 50 percent overstory density. (2) There is a loss in reliability of measurements when forest overstory is assigned to an overstory density class by the spherical densiometer over that of using each measurement directly.

Literature Cited

CRADDOCK, GEORGE W. 1954. Water yield from snow as affected by consumptive water losses. Proc. 22nd Western Snow Conference. Salt Lake City, Utah. April (Processed).

INGEBO, PAUL A. 1955. An instrument for measurement of the density of plant cover over snow course points. Proc. 23rd Western Snow Conference. Portland, Oregon. April (Processed). Discussion by Paul E. Lemmon.

JACKSON, L. W. R. and R. S. HARPER. 1955. Relation of light intensity to basal area of shortleaf pine (*Pinus echinata*) stands in Georgia. Ecology 36: 158-159.

MATUSZ, S. 1953. Swiatlomierz do pomiaru prezepuszczalnosci s w i e t l n e j koron drzew lesnych. (Photometer for measuring the light transmitted through forest tree crowns). Sylwan 97: 367-372.

ROBINSON, MARK W. 1947. An instrument to measure forest crown cover. For. Chron. 23: 222-225.

SUZUKI, T. and T. SATOO. 1954. An attempt to measure the daylight factor under the crown canopy, using a solid angle projecting camera. Bull. Tokyo Univ. For. 46: 169-180. (Jap.) (For. Abst. 16: 2589. 1955).

WEAVER, JOHN E. and F. E. CLEMENTS. 1929. Plant Ecology. McGraw-Hill Company, Inc. New York.

Germination of Slash Pine Pollen in Vitro

BY
R. M. ECHOLS
FRANCOIS MERGEN

FOR MANY SPECIES of plants, tests of pollen germination *in vitro* are used to indicate their pollen viability. Variation in germination percentage may be caused by factors such as drying methods, methods of extraction, conditions during pollen extraction, storage conditions, germination techniques, temperature, relative humidity, and organic and inorganic nature of media (Lidforss, 1899; Brink, 1925; O'Connor, 1927; Dengler and Scamoni, 1939; Smith, 1939, 1942; Saarnljoki, 1941; O'Kelley, 1954; Duffield, 1954). Different tree species show so much variation in chemical requirements for germination of their pollen that a given medium can be considered standard for only a few. Two widely used media are 5 to 10 percent sucrose solutions (or sucrose-agar blocks), and distilled-water (liquid or vapor). In view of the fact that many species respond to varying media combinations, a study of the effect of various inorganic and organic additives on germination of pollen is of interest from both a physiological and cytological point of view.

In this study, the stimulation or inhibition effect of different concentrations of inorganic and organic compounds, the effect of a flower extract, and the effect of a short storage period on the germination of slash pine (*Pinus elliottii* Engelm.) pollen were investigated.

Review of Literature

Investigations into the phenomena of pollen germination and pollen-tube growth were initiated prior to 1900 (Lidfross, 1899). Brink (1925) introduced the hanging drop method of growing pollen *in vitro*. He reported that potassium, sodium, and lithium salts prevented germination at concentrations near M/100, whereas magnesium and barium salts were equally toxic at M/1500. Calcium salts considerably enhanced growth of pollen tubes at concentrations of M/50 to M/500.

Dengler and Scamoni (1939) used sucrose solutions for germination of pollen from twelve tree species. They reported optimum concentrations ranging from 5 percent for *Pinus*, to 40 percent for *Tilia* and other species. The addition of 1 ppm of 3-indoleacetic acid to germination media was found by Smith (1939) to produce favorable effects. He later tested four additional organic compounds and concluded that auxins in concentrations weaker than 20 ppm were favorable additions to a culture medium. All stronger concentrations

A contribution from the John A. Hartford Foundation Program in Forest Biology, Yale School of Forestry, Valhalla, New York. Dr. Mergen is Acting Director of the Research Center. Dr. Echols is now Assistant Professor of Forest Genetics, Arkansas Agricultural and Mechanical College, College Heights, Arkansas.

were toxic, and colchicine caused broadening and rupturing of tubes (Smith, 1942).

Two recent reports of pollen germination tests involved media containing sugar and inorganic salts. Anna F. Faull[1] obtained varied results with sugar series to some of which she had added calcium salts. The calcium salts were not considered effective in altering usual germination patterns. O'Kelley (1954) found that sucrose gave better germination than other sugars, and the addition of a trace of boron seemed to promote pollen tube development.

The effect of living tissue extracts on pollen growth was studied by O'Connor (1927). He concluded that "the fluids of each species of plant and animal contain a simple, diffusible substance of a specific character which is toxic to the protoplasm of all other species, that this substance is not destroyed by boiling, and that it is probably an amino compound." He did not state whether inhibition occurred during germination in extracts from the same plant from which pollen was taken.

Materials and Methods

During the second week of February, 1955, male catkins were collected from four widely separated slash pine trees in the Olustee Experimental Forest, Olustee, Florida. As catkins on outer twigs of the same trees had already started to dehisce naturally, the time was considered optimum for collecting ripe pollen. The catkins were dried in bags made of plastic sausage casing (which was pervious to water vapor), and the pollen was separated with a fine-mesh sieve. During the preparatory and testing period the supply of pollen was stored in vials at 4 degrees C. and 30 percent relative humidity.

Young female strobili at the receptive stage were also collected from the same trees as the male catkins. They were immediately placed in distilled water in the proportion of 25 cones to 50 ml. water. They were allowed to soak overnight to obtain extracts for testing effects on pollen germination.

Germination of the pollen was tested using Brink's hanging drop method as modified by Righter (1939), with the exception of the water-vapor test in which pollen was exposed to 100 percent relative humidity. Double-distilled water was used for a control and for preparation of the following media: iron-complex, 5, 50, and 250 ppm; sucrose, 5 and 10 percent; dextrose, 5 and 10 percent; coconut milk, 10, 25, 50, and 100 percent; thiamine, 10 and 100 ppm; nicotinic acid, 50 and 500 ppm; pyridoxine, 10 and 100 ppm; indoleacetic acid, 1 and 10 ppm; indolebutyric acid, 1 and 10 ppm; naphthaleneacetic acid, 1 and 10 ppm; 2,4-dichlorophenoxy acetic acid, 1 and 10 ppm; 2, 3, 5-triiodobenzoic acid, 1 and 10 ppm; trace element solution; basic mineral solution (Gautheret, 1942); complete mineral nutrient solution (see reference 3, Table 1); ethylenediamine tetraacetic acid, 20 and 200 ppm; and ethylenediamine tetrasodium acetate, 5, 20, and 50 ppm. A total of 220 quadruplicate cultures were made, using 38 different media solutions.

After 96 hours' incubation in a darkened room at 25 degrees C. constant temperature, one hundred pollen grains from each of four trees were counted at random under a magnification of 80 ×. Those which had true pollen tubes longer than the width of the grain were considered to have germinated. Germination percentage for each medium was calculated from total germination out of 400 grains counted.

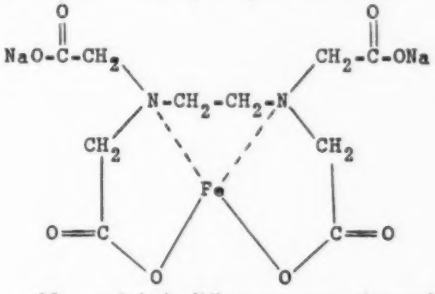
Results

Germination Percentage

Analysis of the germination patterns showed a wide range in response to the media tested, varying from 0 percent to 96 percent (Table 1). Although the standard 10 percent sucrose solution gave 90 percent average germination in the first test, a higher value was observed in all three con-

[1]Paper presented at A. I. B. S. meeting, Gainesville, Fla., Sept. 6, 1954.

centrations of an iron-complex solution. The stock solution was prepared from ethylenediamine tetrasodium acetate as follows: 33.4 gm. of the chemical[2] were dissolved into 356 ml. of distilled water. To this was added 24.9 gm. $Fe_2(SO_4)_3 \cdot H_2O$. The mixture was aerated overnight using a small air pump, then filtered by suction to obtain a stock solution. A dilution of one part stock solution to 200 parts distilled water provided approximately 25 ppm of iron. The following molecular structure represents the iron-complex (Haertl, 1955):



No statistical difference was observed between the two lower concentrations of the iron-complex (50 ppm giving 96 percent germination, and 5 ppm giving 94 percent germination), but the solution containing 250 ppm resulted in only 48 percent germination. This was probably due to the toxic effect of sodium as had been previously reported by Brink (1925). To test whether the stimulation was due to the chelated iron or to the chelation agent, pollen cultures were made in 20 and 100 ppm of ethylenediamine tetrasodium acetate, and 20 and 200 ppm (saturated) ethylenediamine tetraacetic acid solutions. Germination in the tetrasodium media was completely inhibited, while in the latter it was 19 percent for 20 ppm and 0 percent for 200 ppm concentrations. This indicates that the suspended iron in the complex was the stimulating factor for slash pine pollen.

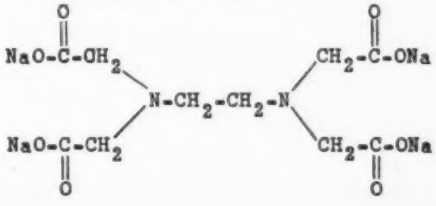
Germination in the control medium (double-distilled water) was 76 percent. In addition to the iron-complex, and the 10 percent sucrose solutions, media which resulted in higher germination were as follows: 5 percent dextrose (84 percent germination), 10 percent dextrose (82 percent), 25 percent coconut milk (78 percent), 10 percent coconut milk (81 percent), 50 ppm nicotinic acid (77 percent), and 1 ppm indolebutyric acid (83 percent germination). Five percent sucrose gave 76 percent germination.

TABLE 1. *Effect of medium composition on germination of slash pine pollen incubated for 96 hours at a temperature of 25 degrees C.*

MEDIUM-ADDITIVE	CONCENTRATION	AVERAGE GERMINATION (%) 0 20 40 60 80 100
Distilled water		
Iron-complex	5 ppm	
"	50 ppm	
"	250 ppm	
Sucrose	5 %	
"	10 %	
Dextrose	5 %	
"	10 %	
Coconut milk	10 %	
"	25 %	
"	50 %	
"	100 %	
Thiamine	10 ppm	
"	100 ppm	
Nicotinic acid	50 ppm	
"	500 ppm	
Pyridoxine	10 ppm	
"	100 ppm	
Indoleacetic acid	1 ppm	
"	10 ppm	
Indolebutyric acid	1 ppm	
"	10 ppm	
Naphthaleneacetic acid	1 ppm	
"	10 ppm	
2,4-dichlorophenoxyacetic acid	1 ppm	
"	10 ppm	
2,3,5-triiodobenzoic acid	1 ppm	
"	10 ppm	
Trace element solution (1)	.1 %	
Basic mineral solution (2)		
Complete mineral nutrient solution (3)		
EDTAA.(4)	20 ppm	
"	200 ppm	
Na4 (5)	5 ppm	
"	20 ppm	
"	50 ppm	
Water vapor		

(1) Refer.: Nitsch, 1951. Full strength.
(2) Basic mineral solution is Knop's Solution, 1/2 strength, with ferric chloride omitted (Gautheret, 1942).
(3) Complete mineral solution prepared by adding 1 ml. of trace element solution (1), plus 1 ml. iron-complex stock solution to 1 liter of basic mineral solution (2).
(4) Ethylenediamine tetraacetic acid.
(5) Ethylenediamine tetrasodium acetate.

[1]This compound is sold commercially under the trade names Sequestrene and Versene and is designated Na4 by one manufacturer. Its chemical structure is as follows:



Response to growth regulators was extremely varied. Indoleacetic acid completely inhibited germination at a concentration of 1 ppm, naphthaleneacetic acid gave 7 percent germination in 1ppm solution, and no germination in 10 ppm solution. Indolebutyric acid at a concentration of 1 ppm resulted in 83 percent germination, while only 57 percent of the pollen germinated at a concentration of 10 ppm.

Germination in coconut milk varied inversely with concentration. With a ten percent dilution in distilled water 81 percent germination was obtained, at a strength of 25 percent it resulted in 78 percent germination, and with a 50 percent dilution 52 percent of the pollen grains germinated. When cultured in 100 percent coconut milk, germination of pollen was only 8 percent. Dense growths of fungi were present in all coconut milk media at the end of the incubation period.

Aqueous extracts from the cones inhibited germination of pollen from all of the trees tested (Fig. 1). When the initial aqueous extracts were diluted with equal volumes of distilled water and used as media, germination of pollen was least inhibited by extracts obtained from cones of the same tree.

The decline in viability of slash pine pollen over a period of 52 days when stored at 4 degrees C. and a relative humidity of 30 percent was tested using the iron-complex solution, 10 percent sucrose, and distilled water (Fig. 2). At the end of this storage period, germination had decreased 16 percent. A statistical analysis showed a significant difference between trees and between germination media, but interaction between storage time and media was not significant. The 16 percent decline in germination was the same, regardless of the medium used for testing.

Nature of Pollen Tubes Produced

Four different types of pollen tubes were found associated with the different media. Most consistent and best developed were the tubes in the iron-complex solution (Figs. 3 and 4). These were without ex-

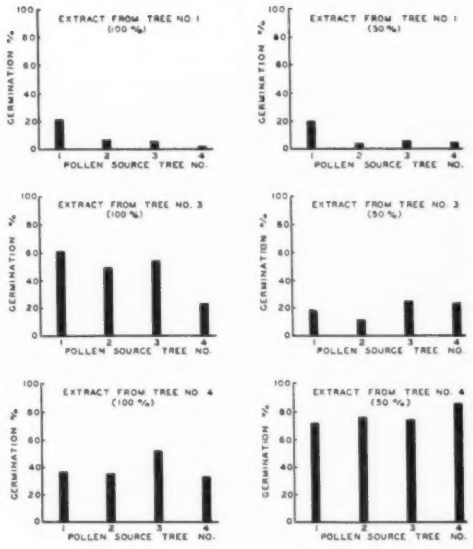

EXTRACT FROM TREE NO. 1 (100 %)
EXTRACT FROM TREE NO. 1 (50 %)
EXTRACT FROM TREE NO. 3 (100 %)
EXTRACT FROM TREE NO. 3 (50 %)
EXTRACT FROM TREE NO. 4 (100 %)
EXTRACT FROM TREE NO. 4 (50 %)
GERMINATION %
POLLEN SOURCE TREE NO.

FIGURE 1. *Germination of slash pine pollen in distilled water containing extracts from young female cones.*

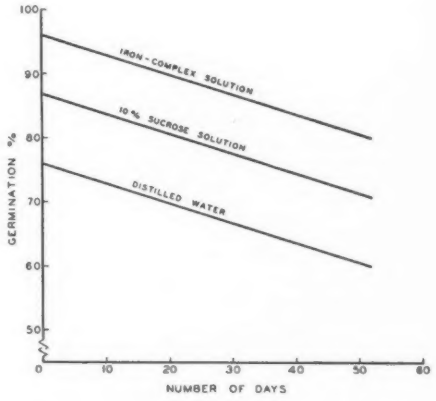


FIGURE 2. *Decrease in viability of slash pine pollen over a period of 52 days while stored at 4 degrees C. and 30 percent relative humidity (tested in three different media).*

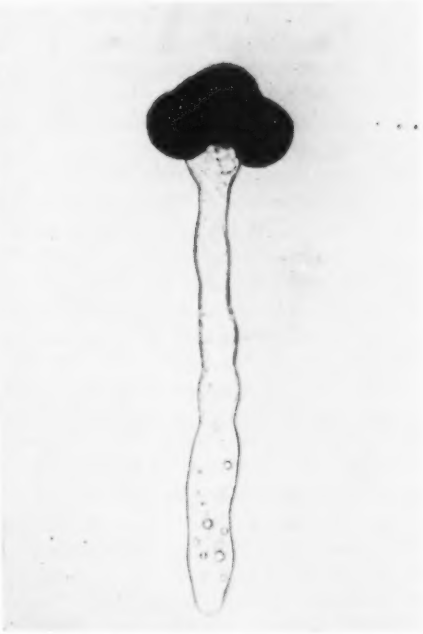


FIGURE 3. *Slash pine pollen grain germinated in the iron-complex solution showing normal development of pollen tube. (× 500)*

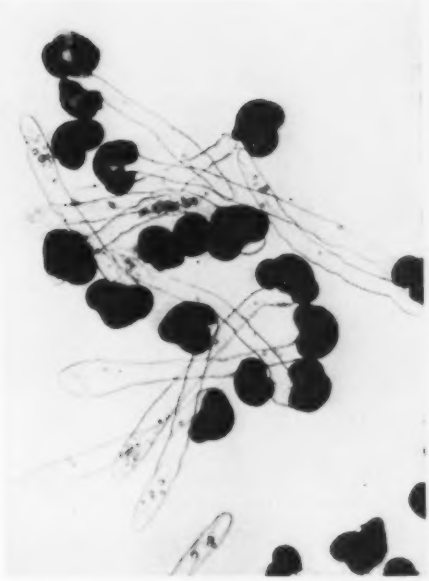


FIGURE 4. *Extent of pollen tube development in the iron-complex solution after an incubation period of 96 hours at a temperature of 25 degrees C. (× 250)*

ception long, intact, and showed the normal internal structures as outlined by Holman and Robbins (1939). Tubes formed in distilled water were also well-developed but generally shorter and of varying lengths. Pollen germinated in 100 percent relative humidity produced medium length pollen tubes, the majority of which were ballooned at the extremities. All sucrose solutions produced short tubes which were filled with closely packed granular bodies, and many showed cytoplasmic extrusion from the tips (Fig. 5). Pollen tubes grown in other media were for the most part very short, abortive or atrophied.

Discussion

Although germination of pollen *in vitro* can be used to indicate potential germination in nature, there are many additional factors, both internal and external, which may limit actual fertilization. This is particularly true for members of the genus *Pinus*. They require a year or longer of growth after germination of pollen takes place before fertilization occurs. Further study is needed to establish the rleation of germination in the laboratory to effective fertilization in nature.

The fluids which are exuded in the micropylar regions and which serve as a vehicle for pollen introduction in pine (Doyle and O'Leary, 1935) may contain substances that not only stimulate germination of pollen but possibly also inhibit growth of fungi. If so, this would indicate a similarity between function of micropylar fluids and the iron-complex solutions used in this study. No fungus mycelia were detected after 96 hours of pollen tube growth in any of the media containing the iron-complex, although all of the sugar solutions became contaminated with fungi during

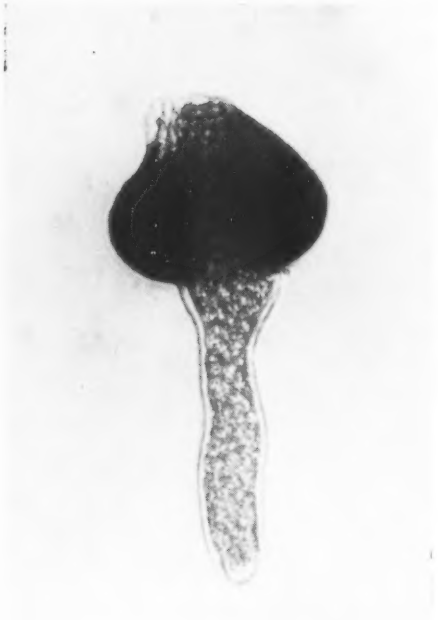


FIGURE 5. *Pollen grain which germinated in a 10 percent sucrose solution. The entire pollen tube is filled with vacuoles. (Photographed under phase contrast illumination* × *780)*

incubation.

The manner in which extractions were made from the young conelets precluded the isolation of physiologically active factors from the fluid in the micropylar openings, because the conelets were completely submerged in distilled water. As a result of this, water soluble substances contained within the cells of the stalk of the conelet were able to diffuse into the extract through the damaged cells near the cut surface. Hormones, or growth regulating substances are present in all tissues of plants, particularly in meristematic cells in the apical regions. From the effect of the growth regulators on the germination of slash pine pollen, we know that their introduction into the extract could either stimulate or inhibit germination and thus influence any direct effect of substances contained in the fluid in the micropylar openings. Therefore, it was not possible to determine whether or not a substance is contained in this germination fluid which inhibits the germination of pollen from the same tree. We know that in slash pine, the seed set is lower in trees that are selfed, and it would be of interest to see whether or not this selection mechanism is active even at the time when pollen grains germinate. It was of interest, however, to observe that the aqueous extracts were physiologically active and that the diluted extracts had a smaller inhibiting effect when tested on pollen collected from the same tree from which the extract had been made. Whether this phenomenon is constant for each tree from year to year is still open for study. It might vary with the physiological state of the trees, and the climatic conditions preceding the period of extraction.

With further study it may be possible to establish within practical limits a direct relationship between percentage of pollen germination and amount of auxin in tissue extracts. Pollen from such species as slash pine or hemlock which retain considerable viability when stored properly may be used for simple auxin tests according to the formula:

$$A = \left[\frac{\%\ \text{germination in control medium}}{\%\ \text{germination in test extract}} - 1 \right] K$$

where A = relative amount of auxin, and K = a conversion factor to be calculated for each type of extract.

Results of this experiment show that external chemicals influence both germination percentage and condition of pollen tubes, and that the two are related. By choice of suitable medium additives, reasonably accurate tests of total germinative capability of pollen may be obtained.

Summary

Germination tests were conducted with pollen from slash pine (*Pinus elliottii*

Engelm.) using 38 different media containing various concentrations of organic and inorganic compounds. Ethylenediamine disodium acetate-iron complex, diluted to contain 5 to 50 ppm of iron produced highest germination (96 percent); 10 percent sucrose resulted in 90 percent germination, whereas only 76 percent of the pollen grains germinated in distilled water. Plant growth regulators gave varying results ranging from complete inhibition to slight stimulation of germination, depending upon concentration in medium. Viability of pollen dropped 16 percent over a period of 52 days while stored at 4 degrees C. and 30 percent relative humidity. Four distinct forms of pollen tubes were found associated with certain media. Aqueous extracts from female flowers inhibited germination of pollen. A formula is suggested for use in a general test for auxins based on relative percentages of pollen germination.

Literature Cited

BRINK, R. A. 1925. The influence of hydrogen-ion concentration on the development of the pollen tube of the sweet pea (*Lathyrus odoratus*). Amer. Jour. Bot. 12: 149-162.

DENGLER, A. and H. SCAMONI. 1939. Uber die Keimungsbedingungen von Waldbaumpollen. Z. Forst-und Jagdw. 71: 1-40.

DOYLE, JOSEPH, and MARY O'LEARY. 1935. Pollination in *Pinus*. Scien. Proc. Roy. Dubl. Soc. 21: 181-190.

DUFFIELD, J. W. 1954. Studies of extraction, storage, and testing of pine pollen. Z. Forstgen. u. Forstpflanz. 3: 39-45.

GAUTHERET, R. J. 1942. Manuel technique de culture des tissues végétaux. Masson et Cie, éditeurs, Boule. Saint-Germain, Paris. 172 pp.

HAERTL, E. J. 1955. New horizons in the application of Chelation to agriculture. Down to Earth (Dow Chemical Co. publ.) 2 (1): 6-9.

HOLMAN, RICHARD M. and WILFRED W. ROBBINS. 1939. A textbook of general botany. John Wiley and Sons, Inc., N. Y. 664 + xvii pp.

LIDFORSS, B. 1899. Weitere Beiträge zur Biologie des Pollens. Jahrb. Wiss. Bot. 33: 232-312.

O'CONNOR, P. 1927. Inhibition of pollen growth by living tissue extracts. Scien. Proc. Roy. Dubl. Soc. 18: 477-484.

O'KELLEY, JOSEPH C. 1955. External carbohydrates in growth and respiration of pollen tubes *in vitro*. Amer. J. Bot. 42: 322-327.

RIGHTER, F. I. 1939. A simple method of making germination tests of pine pollen. J. For. 37: 574-576.

SAARNLJOKI, S. 1941. Experiments on the germination of the pollen of forest trees. Commun. Inst. For. Fenn. 29: 1-15.

SMITH, P. F. 1939. The influence of 3-indole acetic acid on pollen germination. Science 90: 163-164.

SMITH, P. F. 1942. Studies of the growth of pollen with respect to temperature, auxine, colchicine, and vitamin B_1. Amer. J. Bot. 29: 56-66.

Photoperiod and Flowering of Pines

BY
N. T. MIROV

IN SELECTING PINE species for physiological studies such as seed production, it is essential to know how they respond to a changed photoperiod when moved from their natural latitudinal habitat to a more southern or northern locality. Photoperiodic response is understood here in its original sense, that is, the change from a vegetative to a reproductive stage in response to changed length of day (Garner and Allard, 1920). Such behavior should not be confused with the influence of photoperiod on vegetative growth of pines. Several experiments (Kramer, 1937; Vaartaja, 1954; Wareing, 1950) indicate that pine seedlings grow better the longer the daylight period. Vegetative behavior of pines is not the concern of this paper.

Most of the experimental material discussed here is in two places in California: the Institute of Forest Genetics, near Placerville, latitude 38°44′ north, and the University of California, at Berkeley, latitude 37°52′ north. In these two localities less than one degree different latitudinally, the longest days in summer are about 15 hours and the shortest days in winter are about 9½ hours.

Climatically, however, these two places are quite different. The Institute of Forest Genetics is located in the Sierra Nevada at an elevation of about 2,600 feet. The winters at the Institute are characterized by relatively low temperatures: average minimum in January is 34.5° F.; the absolute minimum of record is 11.7° F. Days with temperatures above 100° F. are rather common during July and August. Annual precipitation is 35 inches. Average rainfall in July amounts to 0.01 inch; that of August is zero. The summers are generally cloudless.

At Berkeley, near the Pacific coast, winters are much milder. Average minimum temperature in January is 41.6. Frosts are extremely rare. The lowest temperature ever recorded was 28° F. The annual precipitation of 24 inches is almost entirely in the form of winter rain. The summers are cool and foggy, and the sky is often overcast for days.

Most of the pines of the world are represented at the two places. At the Institute some pines suffer because of the low winter temperatures, lack of rainfall, or perhaps excessive heat and sunlight in the summer. Many of these pines, however, grow well at Berkeley. Erratic behavior of several exotic pines at the Institute, therefore, is caused by factors other than photoperiod.

This paper describes the flowering habits of native and exotic pines planted at the Institute. It also includes some observations

The author is a plant physiologist at the California Forest and Range Experiment Station, Forest Service, U. S. Dept. of Agriculture. He is assigned to the Institute of Forest Genetics, a unit of the station. The work reported here was aided through a grant from Resources for the Future, Inc.

TABLE 1. Flowering of northern pines cultivated at the Eddy Arboretum, Institute of Forest Genetics, Placerville, Calif., 38° 44′ north latitude.[1]

Species	Northern latitude of natural range		Flowering at age 28–30 years	
	From	To	Male flowers[2]	Female flowers
	Degrees			
P. banksiana	41.5	65	abundant	abundant
P. sylvestris var. lapponica[3]	60	70	abundant	abundant
P. sylvestris var. rigensis	57	58	abundant	abundant
P. resinosa	39	52	moderate	moderate
P. montana (several varieties)	40	50	abundant	abundant
P. nigra, several varieties, except mauretanica[4]	40	41	abundant	abundant

[1] *P. cembra* and *P. peuce* are both northern pines; the former is not represented at the Eddy Arboretum; the latter is too young to flower.

[2] Additional information has been published on catkin development and pollen shedding of pines at the Institute (Duffield, 1953).

[3] Seed source: Finland.

[4] Latitudinal range of *P. nigra* var. *mauretanica* is more southern than the latitude of Placerville. This variety also flowers profusely at the Institute.

on the behavior of certain species at Berkeley.

Pines from North of Institute

Most of the species of the genus *Pinus* are found below the 45th parallel in the northern hemisphere. Consequently very few grow wholly north of the Institute. When these were moved south of their natural range, the shorter summer days at Placerville (Fig. 1) did not prevent their producing both male and female flowers (Table 1).

Pinus banksiana, although flowering abundantly at the Institute, does not grow well there. Its poor performance at the Institute is most likely connected with soil conditions, low moisture, and high temperature during summer. Its poor vegetative growth is possibly caused by the short daylight hours, but short days certainly did not prevent flowering of this pine.

All varieties of *Pinus sylvestris* grow very well at the Institute; this pine even reproduces naturally along the irrigation ditch flowing through the arboretum. The flowering performance of *P. sylvestris* var. *lapponica* is more southern than the latitude of Placerville. This variety also flowers profusely at the Institute.

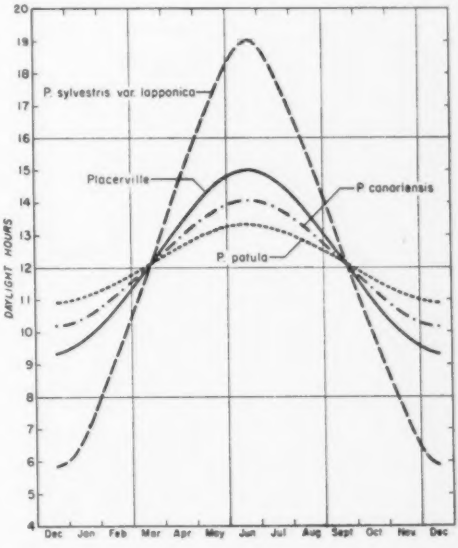


FIGURE 1. *Daylight hours at Placerville, Calif., compared with daylight hours of the natural habitat of three pines:* P. sylvestris *var.* lapponica; P. canariensis, *and* P. patula.

ponica at the Institute is truly surprising.

A similar instance is the normal flowering and reproduction of *Pinus radiata* (habitat 37° to 37.5° north latitude) in southern Ecuador where it was planted at 4° to 5° south latitude late in the nineteenth century. Likewise, *P. radiata*, *P. sylvestris*, and *P. pinaster* develop normally on the equator in Ecuador at about 7,900 feet or 2,400 meters elevation, where they were planted in 1951 (Acosta-Solis, 1954).

Pines that Grow both North and South of Placerville

Pines whose natural habitat lies both north and south of the Institute are numerous. Twenty-four species of this category have been examined at the Eddy Arboretum. Of these pines only four native species exhibit refractory flowering habits. *Pinus albicaulis*, a timberline pine (of the subgenus *Haploxylon*) growing naturally at the same latitude as that of the Institute, has just started to flower at the age of 28 years. *P. lambertiana*, which grows naturally all around the Institute, is also slow in its flowering habits. Although male flowers have been recorded in *P. lambertiana* at the age of 7 years (Righter, 1939), this pine generally does not begin to produce catkins and conelets until about 25 years of age—even then rather sparsely.

Two pinyons, *Pinus monophylla* and *P. edulis*, both pines of an arid environment of the same latitude as the Institute, also reach the reproductive stage rather late. Toward the age of 30 years they have just begun to flower at the Institute.

Causes of the refractory flowering habits of these four native pines are not yet known, but obviously photoperiod is not the cause. The rest of the pines in this category bloom and seed normally at Placerville.

Pinus aristata and *P. balfouriana*, both western high-altitude pines, are not represented at the Eddy Arboretum.

Of interest is the report (Zabelin, 1939) that *Pinus cembroides*, ranging naturally between the 19th and 35th parallels, grows well and produces fertile seed in Crimea, 45° north latitude.

Pines from Southern Latitudes

Pines whose home is in latitudes more southerly than the Institute of Forest Genetics and Berkeley constitute a case of short-day plants exposed to a longer photoperiod during the growing season. Many species of southern latitudes do not survive the low winter temperatures at Placerville, but some from the highlands of Mexico and China withstand the rigors of the Sierra Nevada winters.

Almost all southern pines cultivated at the Institute's arboretum under a longer day condition produce flowers very well (Table 2). *Pinus clausa*, at the age of 27 years, is the only pine in which failure to develop both male and female flowers cannot be explained. In all other southern pines, cases of failure can be traced to factors other than the longer photoperiod. For instance, failure of *P. palustris* to flower is apparently caused by repeated damage inflicted to the trees by wet snow. *P. canariensis* does not flower at the Institute because winter cold nips its tender shoots almost every year. Low winter temperatures most likely are responsible for poor growth and only occasional flowering of *P. torreyana*. At Berkeley, both *P. canariensis* and *P. torreyana* normally develop flowers profusely and set cones every year.

The most striking flowering performance is that of *Pinus patula*. Its home is between 20° and 21° north latitude, in the tropical highland of Mexico at elevations ranging from 5,300 to 9,850 feet. At its natural habitat the longest day is 13½ hours; the shortest is 11 hours (Fig. 1). When cultivated in California, it grows well and flowers profusely (during April) both at the Institute (38°44′) and in Berkeley (37° 52′). Moving this pine almost 20 degrees north has not prevented production of flowers.

TABLE 2. Flowering of southern pines cultivated at Eddy Arboretum, Institute of Forest Genetics, Placerville, Calif., 38° 44′ north latitude.

Species	Latitude of natural range From	To	Flowering habits at age 28–30 years Male flowers	Female flowers
	——*Degrees*——			
P. arizonica	25	34.5	scarce	scarce
P. bungeana	25	35	abundant	abundant
P. canariensis[1]	28	29	none	none
P. cembroides	18	34	moderate	moderate
P. clausa	26.0	30.5	none	none
P. coulteri	28	37	abundant	moderate
P. elliottii	30	33.5	moderate	moderate
P. engelmannii	24	32.5	moderate	scarce
P. excelsa	25	35	moderate	moderate
P. gerardiana	29	35	moderate	moderate
P. glabra	29.5	33.5	light	light
P. greggii	20	26	moderate	moderate
P. leiophylla	17	28	abundant	abundant
P. longifolia	25	35	moderate	scarce
P. luchuensis	29	29.5	moderate	moderate
P. massoniana	22.5	38	moderate	moderate
P. montezumae[2]	15	25	scarce	scarce
P. muricata	35	39	abundant	abundant
P. palustris[3]	27	37	none	none
P. patula	19	21	abundant	abundant
P. quadrifolia	30	33	moderate	moderate
P. radiata	35	37.5	abundant	abundant
P. remorata	29	29.5	abundant	abundant
P. serotina	28	37.5	abundant	abundant
P. taeda	28	38.5	moderate	moderate
P. taiwanensis	22	24	moderate	moderate
P. torreyana[3,4]	33	34	scarce	scarce
P. yunanensis	24	26	moderate	moderate

[1]Repeatedly damaged by cold at the Institute. At Berkeley, Calif. (lat. 37°52′ N.), produces both catkins and conelets in abundance.

[2]Older trees of *P. montezumae* produce cones as far north as 45° north latitude (Zabelin, 1939).

[3]Repeatedly damaged by snow at the Institute.

[4]Older trees of *P. torreyana* at Berkeley produce both catkins and conelets in abundance.

Conclusions

Of course, as Vaartaja (1954) pointed out, different species may vary markedly in their photoperiodic response, and individuals within a species may differ in sensitivity to photoperiod. Perhaps more detailed studies would reveal pines whose flowering is linked with the photoperiod. Thus far, however, no definite connection has been established between photoperiod and the flowering of pines. Of all of the pine species examined—be they from more southern or more northern localities—only *Pinus clausa* failed to flower at the Institute.[1]

In view of these observations, we have arrived at a tentative conclusion: the numerous pines so far inspected performed not as long-day or short-day plants, but as neutral plants whose flowering is not affected by the length of day.

[1]*Pinus clausa* is represented in the Eddy Arboretum by three very poorly developing trees.

The inventory of flowering southern pines at the Institute is admittedly incomplete. Many pines of tropical regions, such as *Pinus merkusii* of Sumatra, *P. insularis* of the Philippines, and *P. tropicalis* of Cuba, are neither represented at the Institute of Forest Genetics' Eddy Arboretum, nor at the same latitude near the coast. Steps have been taken to grow these southern pines at Berkeley where the climate is mild.

Literature Cited

ACOSTA-SOLIS, M. 1954. La forestacion artificial en el Ecuador central. Esc. Politec. Nac. p. 21. Quito.

DUFFIELD, J. W. 1953. Pine pollen collection dates — annual and geographic variation. U. S. For. Serv., Calif. For. Range Exp. Sta. For. Res. Notes 85, 9 pp.

GARNER, W. W., and H. A. ALLARD. 1920. Effect of relative length of day and night and other factors of the environment on growth and reproduction in plants. J. Agric. Res. 18: 553-607.

KRAMER, P. J. 1937. Photoperiodic stimulation of growth by artificial light as a cause of winter killing. Plant Physiol. 12: 881-885.

RIGHTER, F .I. 1939. Early flower production among the pines. J. For. 37: 935-938.

VAARTAJA, OLLI. 1954. Photoperiodic ecotypes of trees. Canad. J. Bot. 32: 392-399.

WAREING, P. F. 1950. Growth studies in woody species I. Photoperiodism in first-year seedlings of *Pinus sylvestris*. Physiol. Plant., Copenhagen 3: 258-276.

ZABELIN, I. A. 1939. Trees and shrubs of the Nikita Botanical Garden. Bull. Nikita Bot. Gard. 22(1): 1-178. Yalta.

FOREST SCIENCE OWNERSHIP STATEMENT

Statement of the ownership, management, etc., required by the Acts of Congress, March 3, 1933 and July 2, 1946, of FOREST SCIENCE, published quarterly at Baltimore, Md., for December, 1956.

DISTRICT OF COLUMBIA

Before me, a Notary Public, Washington, D. C. personally appeared L. Audrey Warren, who having been duly sworn according to law, deposes and says that she is the Business Manager of FOREST SCIENCE and that the following is, to the best of her knowledge and belief, a true statement of the ownership, management, etc., of the aforesaid publication for the date shown in the above caption, required by the Act of March 3, 1933, embodied in section 537, Postal Laws and regulations, printed on the reverse side of this form, to wit:

1. That the names and address of the publisher, editor, and business manager are: publisher, Society of American Foresters, Mills Building, Washington 6, D. C.; editor, Stephen H. Spurr, School of Natural Resources, University of Michigan, Ann Arbor, Michigan; business manager, Miss L. Audrey Warren, Mills Building, Washington 6, D. C.

2. That the owner is the Society of American Foresters, Mills Building, Washington 6, D. C. Stockholders owning 1 per cent or more of total amount of stock are: None.

3. That the known bondholders, mortgagees, and other security holders owning or holding 1 per cent or more of total amount of bonds, mortgages or other securities are: None.

4. That the two paragraphs next above, giving the names of the owners, stockholders and security holders, if any, contain not only the list of stockholders and security holders as they appear upon the books of the company but also, in case where the stockholders or security holders appear upon the books of the company as trustee or in any fiduciary relation, the names of the person or corporation for whom the trustee is acting, is given; also that the said two paragraphs contain statements embracing affiant's full knowledge and belief as to the circumstances and conditions under which stockholders and security holders who do not appear upon the books of the company as trustees, hold stock and securities in a capacity other than that of a bonafide owner; and this affiant has no reason to believe that any other person, association, or corporation has any interest direct or indirect in the paid stock, bonds, or other securities than as so stated by her.

L. AUDREY WARREN, *Business Manager*

Sworn to and subscribed before me this 3rd day of October, 1956 (seal) Henry W. Syfrig, Notary Public, Washington, D. C. (My Commission expires January 14, 1959.)

INDEX TO VOLUME 2, FOREST SCIENCE, 1956

Art.—article; rev.—review; art. (rev.)—review of article or book by.
Authors are indicated by capitals and scientific names by italics.

Abies balsamea (L.) art. 92, 174
Allyl alcohol on growth of Monterey pine seedlings. Transmitted effect of. art. 58
Anthocyanin and leuco-anthocyanins of sugar pine seedlings, The. art. 273
Appalachians. Site index studies of upland oaks in the northern. art. 162
Argentine woods and forests. rev. 285
Arsenite to girdled trees relative to bark peeling. Tissue differentiation and the application of sodium. art. 297

BAILEY, IRVING W. rev. 173
Balsam fir. art. 174
Balsam fir tree volume in the Lake State. art. 92
Bark peeling. Tissue differentiation and the application of sodium arsenite to girdled trees relative to. art. 297
BAVER, L. D. rev. 17
BEAN, JAMES L. art. 187
Beetle. Effects of temperature upon the activity and the behavior of the Douglas fir. art. 258
Big game populations in the National Forests, 1921 to 1950. art. 7
BOYCE, JOHN S., JR. art. 2
BRUCE, DAVID. art. 31
Budworm attack. Pathological aspects of severe spruce. art. 174
Burning on some physical soil properties. Effect of slash. art. 18

CALLAHAM, ROBERT Z. art. 101
CARMEAN, WILLARD H. art. 242
Cenangium dieback fungus. Taxonomy of the. art. 281
Choristoneura fumiferana (Chem.) art. 174
Colchicine. rev. 120
Controlled pollination techniques for the Douglas fir. art. 251
Contributions to plant anatomy. rev. 173
CRAM, W. H. art. 26
Czeckoslovakia. Forestry and wildlife atlas for. rev. 257

Damping-off of poplar seedlings. Prevention of. art. 54
DANA, SAMUEL TRASK. rev. 250
Defoliation on growth of longleaf pine seedlings. Effect of. art. 31
Dendroctonus pseudotsugae Hopk. art. 258
Densiometer for estimating forest overstory density. A spherical. art. 314
DILS, ROBERT E. art. (rev.). 99
Douglas fir beetle. Effects of temperature upon the activity and the behavior of the. art. 258
Douglas fir. Controlled pollination techniques for the. art. 251
Douglas-fir site curves for certain soils in southwestern Washington. Suggested modifications of the. art. 242
Douglas-fir. Windthrow around staggered settlings in old-growth. art. 60
DUSTIN, PIERRE, JR. rev. 120

ECHOLS, R. M. art. 321
EIGSTI, O. J. rev. 120
Effect of defoliation on growth of longleaf pine seedlings. art. 31
Effect of slash burning on some physical soil properties. art. 18
Effect of soaking position on treatability of pine fence posts. art. 43
Effects of temperature upon the activity and the behavior of the Douglas fir beetle. art. 258
Effect on streamflow of cutting a forest understory. art. 82
Endoconidophora fagacearum Bretz. art. 2
ENGLERTH, GEORGE H. art. 2
Experimental design: theory and application. rev. 186

FEDERER, WALTER T. rev. 186
Fence posts. Effect of soaking position on treatability of pine. art. 43
FERCHAU, HUGO A. art. 281
Finland. Growth of forest plantations on different soils of. art. 121
Foliage symptoms as indicators of potassium-deficient soils. art. 113
FORBES, REGINALD D. rev. 22
Forest and range policy. rev. 250
Forest dictionary. note. 126
Forest policy in Scandinavia. rev. 127
Forestry and wildlife atlas for Czechoslovakia. rev. 257
Forestry handbook. rev. 22

German silvicultural systems. art. 75
Germination of slash pine pollen in vitro. art. 321
GILL, TOM. art. (rev.) 285
Girdled trees relative to bark peeling. Tissue differentiation and the application of sodium arsenite to. art. 297
GODWIN, PAUL A. art. 187
GRAHAM, SAMUEL A. art. 132
GRATKOWSKI, H. J. art. 60
Growth of forest plantations on different soils of Finland. art. 121

HETRICK, L. A. art. 181
HOLMSGAARD, ERIK. rev. 105

JOHNSON, EDWARD A. art. 82
JOHNSON, T. W., JR., art. 281

KOVNER, JACOB L. art. 82
KRUGMAN, STANLEY. art. 273
KUNTZ, J. E. art. 54

Land use on a regional basis. Optimum intensity of forest. art. 199
Larch sawfly in the Lake States, The. art. 132
Larix laricina (Du Roi) K. Koch. art. 132
LEAF, ALBERT L. art. 121
Leaves of *Pinus ponderosa*. Variations in copper, boron, and manganese in. art. 190
LEMMON, PAUL E. art. 314
Life history studies of five species of Neodiprion sawflies. art. 181
LINDSEY, ALTON A. art. 287
Longevity of the oak wilt fungus in red oak lumber. art. 2
Longleaf pine seedlings. Effect of defoliation on growth of. art. 31

MARSH, R. E. rev. 127
Maturity of Colorado spruce cones. art. 26
MEEK, W. L. art. 43
MERGEN, FRANCOIS. art. 321
MEYER, ARTHUR B. rev. 22
MIROV, N. T. art. 328

National Forests, 1921 to 1950. Big game populations in. art. 7
Needle oils of three pine species and species hybrids. art. 101
Neodiprion sawflies. Life history studies of five species of. art. 181

Oak lumber. Longevity of the oak wilt fungus in red. art. 2
Oak wilt fungus in red oak lumber. Longevity of. art. 2
Oaks in the northern Appalachians. Site index studies of upland. art. 162
Oils of three pine species and species hybrids. Needle. art. 101
Optimum intensity of forest land use on a regional basis. art. 199
ORR-EWING, A. L. art. 251
OSBORNE, J. G. art. (rev.) 186
Overstory density. A spherical densiometer for estimating forest. art. 314

PARKER, JOHNSON. art. 190
Pathological aspects of severe spruce budworm attack. art. 174
PERSIDSKY, D. J. art. 58
Photoperiod and flowering of pines. art. 328
Picea abies L. art. 121
Picea pungens Engelm. art. 26
Pine fence posts. Effect of soaking position on treatability of. art. 43
Pine forests. Viable seeds from the duff and soil of sugar. art. 36

Pine from juvenile wood and seedling limb sections. Specific gravity estimations of mature loblolly. art. 107
Pine pollen in vitro. Germination of slash. art. 321
Pine, Scotch. art. 121
Pine seedlings. The anthocyanin and leuco-anthocyanins of sugar. art. 273
Pine seedlings. Transmitted effect of allyl alcohol on growth of Monterey. art. 58
Predicting emergence of the white-pine weevil from hibernation. art. 187
Prevention of damping-off of poplar seedlings. art. 54
PRIELIPP, D. O. art. 92
Pristiphora erichsonii Htg. art. 132
Pseudotsuga menziesii (Mirb.) Franco. art. 60, 242, 251, 258

Quercus spp. art. 2, 162
QUICK, CLARENCE R. art. 36

REINES, M. art. 297
RHODES, ROBERT. art. 107
ROTH, ELMER R. art. 2
RUDINSKY, J. A. art. 258

Sampling methods and community attributes in forest ecology. art. 287
Sawflies. Life history studies of five species of Neodiprion. art. 181
Sawfly in the Lake States. The larch. art. 132
SCHANTZ, HOMER L. art. 7
SCHOENIKE, ROLAND. art. (rev.) 120
Seeds from the duff and soil of sugar pine forests. Viable. art. 36
SHEA, KEITH R. art. 54
Silvicultural systems. German. art. 75
Site curves for certain soils in southwestern Washington. Suggested modifications of the standard Douglas-fir. art. 242
Site index studies of upland oaks in the northern Appalachians. art. 162
Slash burning on some physical soil properties. Effect on. art. 18
SMITH, DAVID M. art. (rev.) 22

Pine species and species hybrids. Needle oils of three. art. 101
Pine weevil from hibernation. Predicting emergence of the white. art. 187
Pines. Photoperiod and flowering of. art. 328
Pinus spp. art. 328
Pinus coulteri D. Don. art. 101
Pinus echinata Mill. art. 43
Pinus elliottii Engelm. art. 321
Pinus jeffreyi Grev. and Balf. art. 101
Pinus lambertiana Dougl. art. 273
Pinus palustris Mill. art. 31
Pinus radiata. art. 58
Pinus ponderosa Laws. art. 101
Pinus ponderosa. Variations in copper, boron, and manganese in leaves of. art. 190
Pinus strobus L. art. 187
Pinus sylvestris L. art. 121
Pinus taeda L. art. 107
Pissodes strobi (Peck) art. 187
Plantations on different soils of Finland. Growth of forest. art. 121
Pollen in vitro. Germination of slash pine. art. 321
Pollination techniques for the Douglas fir. Controlled. art. 251
Poplar seedlings. Prevention of damping-off of. art. 54
Populus spp. art. 54
Potassium-deficient soils foliage symptoms as indicators of. art. 113

Soil physics. rev. 17
Soil properties. Effect of slash burning on some physical. art. 18
Specific gravity estimations of mature loblolly pine from juvenile wood and seedling limb sections. art. 107
Spherical densiometer for estimating forest overstory density. A. art. 314
Spruce budworm attack. Pathological aspects of severe. art. 174
Spruce cones. Maturity of Colorado. art. 26
Spruce, Norway. art. 121
SPURR, STEPHEN H. art. (rev.) 17, 250. art. 75

Staggered settings in old-growth Douglas-fir. Windthrow around. art. 60
STAHELIN, R. art (rev.) 257
STEFFERUD, ALFRED. rev. 99
STILLWELL, M. A. art. 174
Streamflow of cutting a forest understory. Effect on. art. 82
STREYFFERT, THORSTEN. art. (rev.) 127
Suggested modifications of the standard Douglas-fir site curves for certain soils in southwestern Washington. art. 242

TARRANT, ROBERT F. art. 18
Taxonomy of the Cenangium dieback fungus. art. 281
Temperature upon the activity and the behavior of the Douglas fir beetle. art. 258
Tissue differentiation and the application of sodium arsenite to girdled trees relative to bark peeling. art. 297
TORTORELLI, LUCAS A. rev. 285
Transmitted effect of allyl alcohol on growth of Monterey pine seedlings. art. 58
Treatability of pine fence posts. Effect of soaking position on. art. 43
Tree-ring analyses of Danish forest trees. rev. 105
TRIMBLE, G. R., JR. art. 162

Variations in copper, boron, and manganese in leaves of *Pinus ponderosa*. art. 190
VITÉ, J. P. art. 258
VOIGHT, G. K. art. 58
Volume in the Lake States. Balsam fir tree. art. 92

WALKER, LAURENCE C. art. 113
WALTERS, C. S. art. 43
Water: yearbook of agriculture, 1955. rev. 99
Weevil from hibernation. Predicting emergence of the white-pine. art. 187
WEITZMAN, SIDNEY. art. 162
WIKSTEN, AKE. art. (rev.) 105
WILCOX, HUGH. art. (rev.) 173
WILDE, S. A. art. 58
Windthrow around staggered settings in old-growth Douglas-fir. art. 60
WINER, HERBERT I. art. (rev.) 22
WORRELL, ALBERT C. art. 199

Yearbook of agriculture, 1955. Water. rev. 99

ZOBEL, BRUCE. art. 107

Suggestions to Contributors

An article submitted for publication should be the best the writer is capable of producing, with all statements, tables, quotations, names and formulas verified before submission. The careful author will have his typescript reviewed by colleagues.

Articles should be typewritten, with 1½-inch margins, on one side only of white bond paper, size 8½ by 11 inches, or 8 by 10½ inches. Carbon copies are not acceptable. All copy should be double-spaced. Pages should be numbered consecutively, preferably in the upper right-hand corner. Only words intended to be set in italics should be underlined.

Material such as equations that exceed 42 spaces on the typewriter cannot be carried in a single line. Such material should either be set so that it can be broken into two or more lines, or should be presented in tabular form.

Tables

Each table should be typewritten on a separate sheet, given a title at the top, and should be numbered consecutively. Even if only one table is submitted, it should be designated Table 1.

Footnotes used in tables should be designated by numerals, never by asterisks or other typographical symbols. These footnotes should be typewritten as part of the table.

Illustrations and Figures

An illustration — whether a photograph, a line drawing, a map, or a graph — is designated as a figure. A caption should be submitted for each illustration, but the caption should be typewritten on a separate sheet.

All figures should be numbered consecutively. If only one is submitted it should be designated Figure 1. On the margin or back of each illustration should be written lightly in soft pencil the number of the figure, the name of the author, and an abbreviated title.

Photographs should be sharp and clear, printed on glossy paper. They should not be smaller than 4 by 5 inches. Sizes 5 by 7 inches and 8 by 10 inches are preferable. Photographs should never be rolled or bent. Care should be taken in fastening photographs with paper clips which often make indentations that show up in reproduction.

Drawings, maps, charts, and graphs are reproduced as line engravings. They should be at least twice as large as they are to appear when reproduced, and should be drawn in india ink on heavy white paper, or tracing cloth.

Footnotes

To indicate a footnote, place a superior figure after the word that refers to the note. Consecutive numerals should be used, never asterisks. Separate the footnote from the text by running a line about one inch inward from the left margin of the type.

Use footnotes to give credit to unpublished material and communications. If only a few references to literature are made (less than one per 1000 words), complete literature citations may be given in footnotes rather than in a separate listing.

Literature Cited

In FOREST SCIENCE practice, references to literature citations are designated by the author's name and year of publication inserted in parentheses at the appropriate place in the text. If there are more than two authors, list only the senior author's name in the text with the abbreviation *et al.* Example: (Smith *et al.*, 1954). Only published references should be given in *Literature Cited*. Periodical abbreviations should follow *Guide to the use of Forestry Abstracts*, Commonwealth Forestry Bureau, Oxford, England, 1950.

Nomenclature and Terminology

When a species is first mentioned in a paper its common name may be immediately followed by its italicized scientific name in parentheses, but this latter name need not be repeated.

As the authority for the exact spelling of tree names, FOREST SCIENCE follows *Check List of Native and Naturalized Trees of the United States (including Alaska)*, Agriculture Handbook 41, Forest Service, U. S. Dept. Agric., Washington, D. C., 1953.

For the spelling of other plant names, contributors should follow *Standardized Plant Names*, 2d Edition, J. Horace McFarland Company, Harrisburg, Pa. 1942.

Technical usage in forestry and allied fields follows *Forestry Terminology*, 2d Edition, Society of American Foresters, Washington 6, D. C. 1950.

There are many style manuals available for the guidance of writers. One of the best is the *Style Manual* of the U. S. Government Printing Office, Washington 25, D. C. 1953.

Webster's *New International Dictionary* is the accepted authority for general spelling.